09/977014 IDS

Part of Paper # 6

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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification [6] **:**
C07K 14/47, 14/52, C12N 15/12, 15/19, 15/63, A61K 38/16, 38/19, 48/00

A1

(11) International Publication Number: **WO 99/29728**

(43) International Publication Date: 17 June 1999 (17.06.99)

(21) International Application Number: PCT/US98/26291

(22) International Filing Date: 11 December 1998 (11.12.98)

(30) Priority Data:
60/069,281 11 December 1997 (11.12.97) US

(63) Related by Continuation (CON) or Continuation-in-Part (CIP) to Earlier Application
US 60/069,281 (CON)
Filed on 11 December 1997 (11.12.97)

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(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: METHOD AND COMPOSITION TO ENHANCE THE EFFICACY OF A VACCINE USING CHEMOKINES

(57) Abstract

The present invention relates to a method to enhance the efficacy of a vaccine in a subject treated with the vaccine comprising administering to the subject in combination with the vaccine a one or more chemokines. The present invention also relates to compositions of vaccines containing chemokines.

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METHOD AND COMPOSITION TO ENHANCE THE EFFICACY OF A VACCINE USING CHEMOKINES

1. CROSS-REFERENCE TO RELATED APPLICATIONS

This application claims priority to U.S. Application Serial No. 60/069,281 filed December 11, 1997.

2. BACKGROUND OF THE INVENTION

The present invention relates to a method to enhance the efficacy of a vaccine by administration of a chemokine, such as macrophage derived chemokine (MDC), in conjunction with the vaccine. The present invention also relates to compositions useful in the method.

2.1. GENERATION OF AN IMMUNE RESPONSE

The introduction of a foreign antigen into an individual elicits an immune response consisting of two major components, the cellular and humoral immune responses, mediated by two functionally distinct populations of lymphocytes known as T and B cells, respectively (see *generally* Coutinho, 1991, Immune System, *Encyclopedia of Human Biology*, Vol. 4, Ed. Dulbecco, Academic Press, Inc.). A subset of T cells responds to antigen stimulation by producing lymphokines which "help" or activate various other cell types in the immune system.

Another T cell subset is capable of developing into antigen-specific cytotoxic effector cells, which can directly kill antigen-positive target cells. On the other hand, the B cell response is primarily carried out by secretory proteins, antibodies, which directly bind and neutralize antigens.

Helper T cells (TH) can be distinguished from classical cytotoxic T lymphocytes (CTL) and B cells by their cell surface expression of the glycoprotein marker CD4. Although the mechanism by which $CD4^+$ TH function has not been fully elucidated, the existence of functionally distinct subsets within the $CD4^+$ T cell compartment has been reported (Mosmann and Coffman, 1989, *Ann. Rev. Immunol.*

7:145-173). In the mouse, type 1 helper T cells (TH1) produce interleukin-2 (IL-2) and τ-interferon (τ-IFN) upon activation, while type 2 helper T cells (TH2) produce IL-4 and IL-5. Based on the profile of lymphokine production, TH1 appear to be involved in promoting the activation and proliferation of other T cell subsets including CTL, whereas TH2 specifically regulate B cell proliferation and differentiation, antibody synthesis, and antibody class switching.

A second T cell subpopulation is the classical CTL which express the CD8 surface marker. Unlike most TH, these cells display cytolytic activity upon direct contact with target cells, rather than through the production of lymphokines. *In vivo*, CTL function is particularly important in situations where an antibody response alone is inadequate. Significant experimental evidence indicates that CTL rather than B cells and their antibody products play a principal role in the defense against viral infections and cancer.

A salient feature of both T and B cell responses is their exquisite specificity for the immunizing antigen; however, the mechanisms for antigen recognition differ between these two cell types. B cells recognize antigens by antibodies, either acting as cell surface receptors or as secreted proteins, which bind directly to antigens on a solid surface or in solution, whereas T cells only recognize antigens that have been processed or degraded into small fragments and presented on a solid phase such as the surface of antigen-presenting cells (APC). Additionally, antigenic fragments must be presented to T cells in association with major histocompatibility complex (MHC)-encoded class I or class II molecules. The MHC refers to a cluster of genes that encode proteins with diverse immunological functions. In man, the MHC is known as HLA. Class I gene products are found on all somatic cells, and they were originally discovered as targets of major transplantation rejection responses. Class II gene products are mostly expressed on cells of various hematopoietic lineages, and they are involved in cell-cell interactions in the immune system. Most importantly, MHC-encoded proteins have been shown to function as receptors for processed antigenic fragments on the surface of APC (Bjorkman et al., 1987, *Nature* 329:506-512).

Another level of complexity in the interaction between a T cell and an antigenic fragment is that it occurs only if the MHC molecules involved are the same on the APC and the responding T cells. In other words, a T cell specific for a particular antigenic epitope expresses a receptor having low affinity for self MHC

proteins, which when such MHC proteins on APC are occupied by the epitope, engage the T cell in a stronger interaction leading to antigen-specific T cell activation. The phenomenon of a T cell reacting with a processed antigen only when presented by cells expressing a matching MHC is known as MHC-restriction.

The specificity of T cell immune responses for antigens is a function of the unique receptors expressed by these cells. The T cell receptor (TCR) is structurally homologous to an antibody; it is a heterodimer composed of disulfide-linked glycoproteins. Four TCR polypeptide chains known as α, β, τ, and δ have been identified, although the vast majority of functional T cells express the αβ heterodimeric TCR. Transfer of α and β genes alone into recipient cells was shown to be both necessary and sufficient to confer antigen specificity and MHC-restriction (Dembic et al., 1986, *Nature* 320:232-238). Thus, the αβ TCR appears to be responsible for recognizing a combination of antigenic fragment and MHC determinants.

The apparent basis of MHC restriction is that $CD4^+$ T cells express αβ TCR which recognize antigenic fragments physically associated with MHC class II proteins, while the TCR on $CD8^+$ CTL recognize MHC class I-associated fragments. Thus, $CD4^+$ T cells can recognize only a restricted class of APC that are class II^+, whereas $CD8^+$ CTL can interact with virtually any antigen-positive cells, since all cells express class I molecules. $CD4^+$ CTL have been identified, and they are MHC class II restricted, and lyse target cells only if the latter express self-MHC class II determinants associated with specific antigenic fragments. Both CD4 and CD8 molecules also contribute to this interaction by binding to monotypic determinants on the MHC class II and I molecules, respectively.

A second type of TCR composed of τδ heterodimers is expressed by a small percentage of T cells, but the involvement of τδ T cells in antigen-specific recognition is still poorly understood. Some studies have shown that functionally active τδ T cells can be cytolytic in a MHC non-restricted manner.

In summary, the generation of an immune response begins with the sensitization of $CD4^+$ and $CD8^+$ T cell subsets through their interaction with APC that express MHC-class I or class II molecules associated with antigenic fragments. The sensitized or primed $CD4^+$ T cells produce lymphokines that participate in the activation of B cells as well as various T cell subsets. The sensitized $CD8^+$ T cells increase in numbers in response to lymphokines and are capable of destroying any

cells that express the specific antigenic fragments associated with matching MHC-encoded class I molecules. For example, in the course of a viral infection, CTL eradicate virally-infected cells, thereby limiting the progression of virus spread and disease development.

2.2. ANTIGEN PRESENTING CELLS

The presentation of antigens to T cells is carried out by specialized cell populations referred to as antigen presenting cells (APC). Typically, APC include macrophages/monocytes, B cells, and bone marrow derived dendritic cells (DC). APC are capable of internalizing exogenous antigens, cleaving them into smaller fragments in enzyme-rich vesicles, and coupling the fragments to MHC-encoded products for expression on the cell surface (Goldberg and Rock, 1992, *Nature* 357:375-379). Since APC express both MHC-encoded class I and class II glycoproteins, they can present antigenic fragments to both $CD4^+$ and $CD8^+$ T cells for the initiation of an immune response.

By definition, APC not only can present antigens to T cells with antigen-specific receptors, but can provide all the signals necessary for T cell activation. Such signals are incompletely defined, but probably involve a variety of cell surface molecules as well as cytokines or growth factors. Further, the factors necessary for the activation of naive or unprimed T cells may be different from those required for the re-activation of previously primed memory T cells. The ability of APC to both present antigens and deliver signals for T cell activation is commonly referred to as an accessory cell function. Although monocytes and B cells have been shown to be competent APC, their antigen presenting capacities *in vitro* appear to be limited to the re-activation of previously sensitized T cells. Hence, they are not capable of directly activating functionally naive or unprimed T cell populations.

Although it had been known for a long time that APC process and present antigens to T cells, it was not shown until relatively recently that small antigenic peptides could directly bind to MHC-encoded molecules (Babbit et al., 1985, *Nature* 317:359; Townsend et al., 1986, *Cell* 44:959). However, it is believed that, normally, complex antigens are proteolytically processed into fragments inside the APC, and become physically associated with the MHC-encoded proteins intracellularly prior to

trafficking to the cell surface as complexes. Two distinct pathways for antigen presentation have been proposed (Braciale et al., 1987, *Immunol. Rev.* 98:95-114). It was thought that exogenous antigens were taken up by APC, processed and presented by the exogenous pathway to class II restricted $CD4^+$ T cells, while the endogenous pathway processed intracellularly synthesized proteins, such as products of viral genes in virally-infected cells, for association with MHC class I proteins and presentation to $CD8^+$ CTL. However, although the two pathways in antigen processing and presentation may still be correct in some respects, the distinction is blurred in light of recent findings that exogenously added antigens may also be presented to class I-restricted CTL (Moore et al., 1988, *Cell* 54:777).

The term "dendritic cells" (DC) refers to a diverse population of morphologically similar cell types found in a variety of lymphoid and non-lymphoid tissues (Steinman, 1991, *Ann. Rev. Immunol.* 9:271-296). These cells include lymphoid DC of the spleen, Langerhans cells of the epidermis, and veiled cells in the blood circulation. Although they are collectively classified as a group based on their morphology, high levels of surface MHC-class II expression, and absence of certain other surface markers expressed on T cells, B cells, monocytes, and natural killer cells, it is presently not known whether they derive from a common precursor or can all function as APC in the same manner. Further, since the vast majority of published reports have utilized DC isolated from the mouse spleen, results from these studies may not necessarily correlate with the function of DC obtained from other tissue types. (Inaba et al., 1997, *J. Exp. Med.* 166:182-194; Hengel et al., 1987, *J. Immunol.*, 139:4196-4202; Kaut et al., 1988, *J. Immunol.*, 140:3186-3193; Romani et al., 1989, *J. Exp. Med.* 169:1169-1178; Macatonia et al., 1989, *J. Exp. Med.* 169:1255-1264; Inaba et al., 1990, *J. Exp. Med.* 172:631-6640). For example, despite high levels of MHC-class II expression, mouse epidermal Langerhans cells, unlike splenic DC, are not active APC in mixed leucocyte reaction (MLR), unless cultured with granulocyte-macrophage colony stimulating factor (GM-CSF) (Witmer-Pock et al., 1987, *J. Exp. Med.* 166:1484-1498; Heufler et al., 1988, *J. Exp. Med.* 167:700-705). Most human Langerhans cells express the CD1 and CD4 markers, while blood DC do not. Additionally, it has not been established the extent to which the functional characteristics observed with mouse DC are applicable to human DC, especially the DC obtained from non-splenic tissues; in part, due to inherent differences between the

human and murine immune systems.

Recently, a few studies have described the isolation of human DC from the peripheral blood, which involves the use of sheep red blood cells and/or fetal calf serum (Young and Steinman, 1990, *J. Exp. Med.* 171:1315-1332; Freudenthal and Steinman, 1990, *Proc. Natl. Acad. Sci. USA* 87:7698-7702; Macatonia et al., 1989 *Immunol.* 67:285-289; Markowicz and Engleman, 1990, *J. Clin. Invest.* 85:955-961). Engleman et al. described a partial purification procedure of DC from human blood, which does not involve the use of sheep red blood cells and/or fetal calf serum, and showed that the partially purified human DC can, in fact, present exogenous antigens to naive T cells (PCT Publication WO 94/02156 dated February 3, 1994 at page 9, lines 5-32).

Recent studies have indicated that DCs are superior APCs as compared to other APCs such as macrophages and monocytes. First, the potent accessory cell function of DCs provides for an antigen presentation system for virtually any antigenic epitopes which T and B cells are capable of recognizing through their specific receptors. For example, Engleman et al. demonstrate that human DCs can present both complex protein antigens and small peptides to $CD4^+$ T cells as well as to as $CD8^+$ CTL (PCT Publication WO 94/02156 dated February 3, 1994, Example 7, from page 29, line 10 to page 34, line 16). Engleman et al. also show that the *in vitro* priming effect of DCs does not require the addition of exogenous lymphokines, indicating that DCs produce all of the necessary signals in antigen presentation leading to the activation of T cells (PCT Publication WO 94/02156 dated February 3, 1994, from page 32, line 36 to page 33, line 2). More importantly, DCs can induce a primary $CD4^+$ T cell-mediated proliferative response when similarly prepared monocytes can not induce such a response (PCT Publication WO 94/02156 dated February 3, 1994 at page 31, lines 23-30). Similarly, when DCs and monocytes ware compared for their ability to present antigens for re-activating secondary T cell response, it was observed that DCs were capable of stimulating a stronger response than monocytes (PCT Publication WO 94/02156 dated February 3, 1994 at page 32, lines 12-16).

2.3. CHEMOKINES

Chemokines, or chemoattractant cytokines, are a subgroup of immune factors

that have been shown to mediate chemotactic and other pro-inflammatory phenomena (see, Schall, 1991, *Cytokine* 3:165-183). Chemokines are small molecules of approximately 70-80 residues in length and can generally be divided into two subgroups, α which have two N-terminal cysteines separated by a single amino acid (CxC) and β which have two adjacent cysteines at the N terminus (CC). RANTES, MIP-1α and MIP-1β are members of the β subgroup (reviewed by Horuk, R., 1994, *Trends Pharmacol. Sci.* 15:159-165; Murphy, P.M., 1994, *Annu. Rev. Immunol.* 12:593-633; Baggiolini et al. *Annu. Rev. Immunol.* 1997, 15:675-705).

MCP-1 has been shown to attract monocytes but not neutrophils. MCP-1, MCP-2, and MCP-3 share a pyroglutamate proline NH_2-terminal motif and are structurally closely related to each other and to eotaxin (56% to 71% amino acid sequence identity). MCP-1, MCP-2, and MCP-3 attract monocytes, $CD4^+$ and $CD8^+$ T lymphocytes (Loetscher et al. *FAESB J.* 1994, 8:1055-60), as well as basophil leukocytes. MCP-2, MCP-3, and MCP-4 (but not MCP-1) attracts eosinophil leukocytes. All four MCPs attract activated T lymphocytes, natural killer (NK) cells, and dendritic cells (see Baggiolini et al. *Annu. Rev. Immunol.* 1997, 15:675-705).

Eotaxin acts on eosinophils and is inactive on neutrophils and monocytes, but has weak-to-moderate chemotactic activity toward IL-2-conditioned T lymphocytes (see Baggiolini et al. *Annu. Rev. Immunol.* 1997, 15:675-705). Due to its preferential, powerful action on eosinophils and its occurrence in different species, eotaxin is considered to be an important chemokine in the pathophysiology of allergic conditions and asthma (See Baggiolini et al. *Annu. Rev. Immunol.* 1997, 15:675-705).

IP10 is a CXC chemokine attracts human monocytes, T lymphocytes, and NK cells, and Mig attracts tumor-infiltrating T lymphocytes. It has been suggested that IP10 and Mig may also be involved in the regulation of lymphocyte recruitment and the formation of the lymphoid infiltrates observed in autoimmune inflammatory lesions, delayed-type hypersensitivity, some viral infections, and certain tumors (Baggiolini et al. *Annu. Rev. Immunol.* 1997, 15:675-705).

SDF-1 (stromal cell–derived factor 1), including SDF-1 and SDF-1ß stimulates the proliferation of B cell progenitors, and attracts mature dendritic cells (Finkel et al. *Immunobiology* 1998, 198:490-500). Synthetic human SDF-1 stimulates monocytes, neutrophils, and peripheral blood lymphocytes, as is indicated by $[Ca2+]i$ changes and chemotaxis. SDF-1 is also a powerful HIV-suppressive factor (See Baggiolini et al.

Annu. Rev. Immunol. 1997, 15:675-705).

The amino terminus of the β chemokines RANTES, MCP-1, and MCP-3 has been implicated in the mediation of cell migration and inflammation induced by these chemokines. This involvement is suggested by the observation that the deletion of the amino terminal 8 residues of MCP-1, amino terminal 9 residues of MCP-3, and amino terminal 8 residues of RANTES and the addition of a methionine to the amino terminus of RANTES, antagonize the chemotaxis, calcium mobilization and/or enzyme release stimulated by their native counterparts (Gong et al., 1996, *J. Biol. Chem.* 271:10521-10527; Proudfoot et al., 1996 *J. Biol. Chem.* 271:2599-2603). Additionally, α chemokine-like chemotactic activity has been introduced into MCP-1 via a double mutation of Tyr 28 and Arg 30 to leucine and valine, respectively, indicating that internal regions of this protein also play a role in regulating chemotactic activity (Beall et al., 1992, *J. Biol. Chem.* 267:3455-3459).

The monomeric forms of all chemokines characterized thus far share significant structural homology, although the quaternary structures of α and β groups are distinct. While the monomeric structures of the β and α chemokines are very similar, the dimeric structures of the two groups are completely different. An additional chemokine, lymphotactin, which has only one N terminal cysteine has also been identified and may represent an additional subgroup (γ) of chemokines (Yoshida et al., 1995, *FEBS Lett.* 360:155-159; and Kelner et al., 1994, *Science* 266:1395-1399).

Receptors for chemokines belong to the large family of G-protein coupled, 7 transmembrane domain receptors (GCR's) (*See*, reviews by Horuk, R., 1994, *Trends Pharmacol. Sci.* 15:159-165; and Murphy, P.M., 1994, *Annu. Rev. Immunol.* 12:593-633). Competition binding and cross-desensitization studies have shown that chemokine receptors exhibit considerable promiscuity in ligand binding. Examples demonstrating the promiscuity among β chemokine receptors include: CCR-1, which binds RANTES and MIP-1α (Neote et al., 1993, *Cell* 72:415-425), CCR-4, which binds RANTES, MIP-1α, and MCP-1 (Power et al., 1995, *J. Biol. Chem.* 270:19495-19500), and CCR-5, which binds RANTES, MIP-1α, and MIP-1β (Alkhatib et al., 1996, *Science* 272:1955-1958 and Dragic et al., 1996, *Nature* 381:667-674). Erythrocytes possess a receptor (known as the Duffy antigen) which binds both α and β chemokines (Horuk et al., 1994, *J. Biol. Chem.* 269:17730-17733; Neote et al., 1994, *Blood* 84:44-52; and Neote et al., 1993, *J. Biol. Chem.* 268:12247-12249). Thus the sequence and

structural homologies evident among chemokines and their receptors allow some overlap in receptor-ligand interactions.

Godiska et al. identified and described the nucleic acid and amino acid sequences of an additional β chemokine designated macrophage derived chemokine (MDC) (PCT Publication WO 96/40923 dated December 19, 1996, and 1997, *J. Exp. Med.* 185:1595-1604). PCT publication WO 96/40923 further provides materials and methods for the recombinant production of the chemokine, the purified and isolated chemokine protein, and polypeptide analogues thereof. The PCT publication WO 96/40923 does not disclose that the human MDC has chemotactic activity upon DC. While Godiska et al. (1997, *J. Exp. Med.* 185:1595-1604) showed that, in a microchamber migration assay, monocyte-derived DC migrated toward the human MDC, the reference fails to teach that MDC can enhance an immune response to an antigen *in vivo*.

Chang et al. (1997, *J. Biol. Chem.* 272(40):25229-25237), isolated a stimulated T cell chemotactic protein (STCP-1) from an activated macrophage cDNA library. The nucleotide sequence of the STCP-1 is identical to that of the MDC isolated by Godiska et al. (PCT Publication WO 96/40923 dated December 19, 1996, and 1997, *J. Exp. Med.* 185:1595-1604). However, unlike the results observed by Godiska et al. (1997, *J. Exp. Med.* 185:1595-1604), Chang et al. (1997, *J. Biol. Chem.* 272(40):25229-25237) showed that although the STCP-1 acted as a mild chemoattractant for primary activated T lymphocytes and a potent chemoattractant for chronically activated T lymphocytes, the STCP-1 has no chemoattractant activity for monocytes, neutrophils, eosinophils and resting T lymphocytes. Chang et al. further showed that the STCP-1 does not induce $Ca2^{+}$ mobilization in monocytes, dendritic cells, neutrophils, eosinophils, lipopolysaccharide-activated B lymphocytes, and freshly isolated resting T lymphocytes.

2.4. HIV VACCINES

Human immunodeficiency virus (HIV) induces a persistent and progressive infection leading, in the vast majority of cases, to the development of the acquired immunodeficiency syndrome (AIDS) (Barre-Sinoussi et al., 1983, *Science* 220:868-870; Gallo et al., 1984, *Science* 224:500-503). The HIV envelope surface glycoproteins are

synthesized as a single 160 kilodalton precursor protein which is cleaved by a cellular protease during viral budding into two glycoproteins, gp41 and gp120. gp41 is a transmembrane glycoprotein and gp120 is an extracellular glycoprotein which remains non-covalently associated with gp41, possibly in a trimeric or multimeric form (Hammerskjold, M. and Rekosh, D., 1989, *Biochem. Biophys. Acta* 989:269-280). The V3 loop of gp120 is the major determinant of sensitivity to chemokine inhibition of infection or replication (Cocchi et al., 1996, *Nature Medicine* 2:1244-1247; and Oravecz et al., 1996, *J. Immunol.* 157:1329-1332).

Although considerable effort is being put into the design of effective therapeutics, currently no curative anti-retroviral drugs against AIDS exist. The HIV-1 envelope proteins (gp160, gp120, gp41) have been shown to be the major antigens for neutralizing anti-HIV antibodies present in AIDS patients (Barin et al., 1985, *Science* 228:1094-1096). Thus far, therefore, these proteins seem to be the most promising candidates to act as antigens for anti-HIV vaccine development. Several groups have begun to use various portions of gp160, gp120, and/or gp41 as immunogenic targets for the host immune system (see, for example, Ivanoff et al., U.S. Pat. No. 5,141,867; Saith et al., PCT publication WO 92/22654; Shafferman, A., PCT publication WO 91/09872; Formoso et al., PCT publication WO 90/07119). Therefore, methods to increase the efficacy of vaccines against HIV, especially vaccines using gp120 as the antigen, are needed.

Additionally a novel vaccine technology, designated genetic vaccination, nucleic acid vaccination or DNA vaccination, has been explored to induce immune responses *in vivo*. Injection of cDNA expression cassettes results in *in vivo* expression of the encoded proteins (Dubensky et al., 1984, *Proc. Natl. Acad. Sci. USA* 81:7529-7533; Raz et al., 1993, *Proc. Natl. Acad. Sci. USA* 90:4523; Wolff et al., 1990, *Science* 247:1465-1468), with the concomitant development of specific cellular and humoral immune responses directed against the encoded antigen(s) (Wang et al., 1995, *Hum. Gene Ther.* 6:407-418; Ulmer et al., 1993, *Science* 259:1745-1749; Tang et al., 1992, *Nature* 356:152-154; Michel et al., 1995, *Proc. Natl. Acad. Sci. USA* 92:5307-5311; and Lowrie et al., 1994, *Vaccine* 12:1537-1540). Humoral and cellular responses have been induced to HIV-1 and SIV antigens through various applications of this technology in macaques (Wang et al., 1995, *Virology* 221:102-112; Wang et al., 1993, *Proc. Natl. Acad. Sci. USA* 90:4156-4160; and Boyer et al., 1996, *J. Med.*

Primatol. 25:242-250) as well as mice (Wang et al., 1995, *Virology* 221:102-112; Lu et al., 1995, *Virology* 209:147-154; Haynes et al., 1994, *AIDS Res. Hum. Retroviruses* 10 (Suppl. 2):S43-S45; Okuda et al., 1995, *AIDS Res. Hum. Retroviruses* 11:933-943).

Recently, Lekutis et al. (1997, *J. Immunol.* 158:4471-4477), assessed the TH cell response elicited by an HIV-1 gp120 DNA vaccine in rhesus monkeys by isolation of gp120-specific, MHC class II-restricted CD4^{+} T cell lines from the vaccinated animals. Lekutis et al. showed that the isolated cell lines proliferated in response to APC in the presence of recombinant gp120, as well as to APC expressing HIV encoded env protein. Lekutis et al. further showed that these cell lines responded to env by secreting IFN-Γ and IFN-α without appreciable IL-4 production. These results demonstrate that the animals exhibited a cellular immune response to the DNA vaccine.

Boyer et al. (1997, *Nature Medicine* 3:625-532), inoculated chimpanzees with an HIV-1 DNA vaccine encoding env, rev, and gag/pol, and found that the immunized animals developed specific cellular and humoral immune responses to these proteins. After challenging the immunized animals with a heterologous chimpanzee titered stock of HIV-1 SF2, Boyer et al. further found, using a Reverse Transcriptase-Polymerase Chain Reaction (RT-PCR) assay, that those animals vaccinated with the DNA vaccine were protected against infection whereas the control animals were not so protected.

Kim et al., (1997 *J. Immunol.* 158:816-826), investigated the role of co-delivery of genes for IL-12 and GM-CSF along with DNA vaccine formulation for HIV-1 antigens env and gag/pol in mice. Kim et al. observed a dramatic increase in specific CTL response from the mice immunized with the HIV-1 DNA vaccine and IL-12. Kim et al. also observed that the co-delivery of IL-12 genes resulted in the reduction of specific antibody response, whereas the codelivery of GM-CSF genes resulted in the enhancement of specific antibody response. Kim et al. further observed that co-delivery of IL-12 gene with a HIV DNA vaccine results in splenomegaly (Kim et al. 1997, *J. Immunol.*, 158:816-826), which has been shown in mice to have toxic effects such as weight reduction or even death (Eng et al., 1995, *J. Exp. Med.* 181:1893; Stevensen et al., 1995, *J. Immunol.* 155:2545; and Orange et al., 1995, *J. Exp. Med.* 181:901).

Notwithstanding the recent developments of the HIV DNA vaccine, there still

exists a need for a method to enhance the efficacy of a vaccine, especially an HIV DNA vaccine. For instance, for efficacious vaccine against HIV-1 one preferably induces both cellular and humoral immune responses to control the infection (Boyer et al., 1997, *Nature Medicine* 3:625-532). The induction of both cellular and humoral immune response by the Berjer et al. method is still quite low because only one of the three immunized chimpanzees developed both cellular and humoral responses. Similarly, although co-delivery of an IL-12 encoding gene with a HIV DNA vaccine, as described in Kim et al. (1997, *J. Immun.* 158:816-826), may have enhanced the cellular immune response, this co-delivery also decreased the humoral response.

Citation of a reference hereinabove shall not be construed as an admission that such reference is prior art to the present invention.

3. SUMMARY OF THE INVENTION. SUMMARY OF THE INVENTION. . SUMMARY OF THE INVENTION

The present invention is based upon the ability of chemokines, such as MDC, Rantes, MIP-1d, MIP-1B, and I-309, to enhance the immune response to an antigen, particularly a vaccine. Accordingly, in a first aspect, the present invention provides a method for enhancing the efficacy of a vaccine, which method comprises administration to a subject of one or more purified chemokines, or biologically active fragments, analogues or derivatives thereof, either concurrently with one or more purified antigens against which an immune response is desired or within a time period either before or after administration of the antigens such that the immune response against the antigens is enhanced.

In a second aspect, the present invention provides a method to enhance the efficacy of a vaccine, which method comprises administration to a subject of a first set of one or more purified nucleic acids comprising one or more nucleotide sequences encoding one or more chemokines, or fragments, derivatives, analogues, and/or truncation isoforms thereof, and a second purified nucleic acid comprising a nucleotide sequence encoding one or more antigens against which an immune response is desired, such that, the one or more chemokine(s) and the antigen(s) are expressed in a coordinated manner upon introduction into a suitable cell. Alternatively, the nucleotide sequences encoding one or more chemokines, or

fragments, derivatives, and/or analogues thereof, and the antigens against which an immune response is desired are present on the same nucleic acid.

In a preferred embodiment, the invention provides a method to enhance the efficacy of an HIV vaccine.

In yet another aspect, the present invention provides a composition comprising an immunogenic amount of one or more purified antigens, an amount of one or more purified chemokines, or a fragments, derivatives, analogues and/or truncation isoforms thereof, effective to enhance the immune response to the antigen. In another aspect, the present invention provides a composition comprising a first set of one or more purified nucleic acids comprising one or more nucleotide sequences encoding one or more chemokines, fragments, derivatives analogues and or truncation isoforms thereof, and a second set of purified nucleic acids comprising one or more nucleotide sequences encoding one or more antigens against which an immune response is desired, such that, the chemokine(s) and the antigen are expressed in a coordinated manner upon introduction into a suitable cell. In a preferred embodiment, the antigen is an HIV antigen. In another preferred embodiment, the chemokine is selected from the group consisting of: Macrophage-derived chemokine, Monocyte chemotactic protein 1, Monocyte chemotactic protein 2, Monocyte chemotactic protein 3, Monocyte chemotactic protein 4, activated macrophage specific chemokine 1, Macrophage inflammatory protein 1 alpha, Macrophage inflammatory protein 1 beta, Macrophage inflammatory protein 1 gamma, Macrophage inflammatory protein 1 delta, Macrophage inflammatory protein 2 alpha, Macrophage inflammatory protein 3 alpha, Macrophage inflammatory protein 3 beta, Regulated upon activation, normal T cell expressed and secreted (and its variants), I-309, EBI1-ligand chemokine, Pulmonary and activation regulated chemokine, Liver and activation-regulated chemokine, Thymus and activation regulated chemokine, Eotaxin (and variants), Human CC chemokine 1, Human CC chemokine 2, Human CC chemokine 3, IL-10-inducible chemokine, liver-expressed chemokine, 6Ckine, Exodus 1, Exodus 2, Exodus 3, thymus-expressed chemokine, Secondary Lymphoid tissue chemokine, Lymphocyte and Monocyte chemoattractant; Monotactin, Activation-induced, chemokine-related molecule, Myeloid progenitor inhibitory factor-1, Myeloid progenitor inhibitory factor-2, Stromal cell-derived factor 1 alpha, Stromal cell-derived factor 1 beta, B-cell-attracting chemokine 1, HuMIG, H174, Interferon-stimulated T-cell alpha

chemoattractant, Interleukin-8, IP-10, platelet factor 4, growth-regulated gene-alpha, growth-regulated gene-beta, growth-regulated gene-gamma, Neutrophil-activating protein 2, ENA-78, granulocyte chemotactic protein 2, LYMPHOTACTIN, and Fractalkine/neurotactin.

4. DESCRIPTION OF FIGURES

Figures 1A and 1B. The nucleotide and amino acid sequences of MDC. 1A depicts the nucleotide sequence of MDC (SEQ ID NO:1), with the coding region indicated by the appearance of the amino acid sequence in the line below; and 1B depicts the amino acid of MDC (SEQ ID NO:2) from GenBank accession no. U83171 (Godiska et al., 1997, *J. Exp. Med.* 185:1595-1604).

5. DETAILED DESCRIPTION OF THE INVENTION

The present invention relates to a method for enhancing the efficacy of a vaccine in a subject comprising administering to the subject one or more purified antigens in conjunction with one or more purified chemokines, or more purified fragments, derivatives or analogues and/or truncation isoforms thereof.

While any chemokine may be employed according to the present invention, the chemokine is preferably selected from the following table:

Chemokine Class	Chemokines	Abbreviations	Accession Number
CC Chemokines	Macrophage-derived chemokine	MDC/STCP-1	u83171; u83239
	Monocyte chemotactic protein 1	MCP-1	x14768
	Monocyte chemotactic protein 2	MCP-2	X99886
	Monocyte chemotactic protein 3	MCP-3	x72308; s57464
	Monocyte chemotactic protein 4	MCP-4	u46767
	activated macrophage specific chemokine 1	AMAC-1	Y13710
	Macrophage inflammatory protein 1 alpha	MIP-1α	AF043339; X03754; D90144

Chemokine Class	Chemokines	Abbreviations	Accession Number
CC Chemokines (continued)	Macrophage inflammatory protein 1 beta	MIP-1β	j04130; d90145
	Macrophage inflammatory protein 1 gamma	MIP-1γ	
	Macrophage inflammatory protein 1 delta	MIP-1δ	AF031587
	Macrophage inflammatory protein 2 alpha	MIP-2α	AF043340
	Macrophage inflammatory protein 3 alpha	MIP-3α	u77035
	Macrophage inflammatory protein 3 beta	MIP-3β	u77180
	Regulated upon activation, normal T cell expressed and secreted (and its variants)	RANTES	M21211
	I-309		M57502
	EBI1-ligand chemokine	ELC	AB000887
	Pulmonary and activation regulated chemokine	PARC/DC-CK-1/MIP4	AB000221
	Liver and activation-regulated chemokine	LARC	D86955
	Thymus and activation regulated chemokine	TARC	D43767
	Eotaxin (and variants)		D49372; Z69291; Z75669; Z75668
	Human chemokine 1	HCC1; NCC2	Z49270; z49269
	Human chemokine 2	HCC2; NCC3, MIP-5, MIP-1δ	Z70292
	Human chemokine 3	HCC3	Z70293
	IL-10-inducible chemokine	HCC4	U91746
	liver-expressed chemokine.	LEC; HCC4;NCC4	AB007454
	6Ckine		AF001979
	Exodus 1		u64197
	Exodus 2		U88320
	Exodus 3		U88321
	thymus-expressed chemokine	TECK	U86358
	Secondary Lymphoid tissue chemokine	SLC	AB002409

Chem kine Class	Chem kines	Abbreviations	Accession Number
CC Chemokines (continued)	Lymphocyte and Monocyte chemoattractant; Monotactin	LMC	AF055467
	Activation-induced, chemokine-related molecule	ATAC	x86474
	Myeloid progenitor inhibitory factor-1	MPIF-1; MIP-3 or ckbeta8	u85767
	Myeloid progenitor inhibitory factor-2	MPIF-2	u85768
	Stromal cell-derived factor 1 alpha	SDF-1α; PBSF	L36034
CXC chemokines	Stromal cell-derived factor 1 beta	SDF-1β; PBSF	L36033
	B-cell-attracting chemokine 1	BLC	AJ002211
	HuMIG		x72755 s60728
	H174		AF002985
	Interferon-stimulated T-cell alpha chemoattractant	I-TAC	AF030514
	Interleukin-8	IL-8	m17017; y00787
	IP-10		X02530
	platelet factor 4	PF4	M20901
	growth-regulated gene-alpha	GRO-α	J03561
	growth-regulated gene-beta	GRO-β	M36820
	growth-regulated gene-gamma	GRO-γ	M36821
	Neutrophil-activating protein 2	NAP-2; CTAP-3	M54995; M38441
	ENA-78		L37036
	granulocyte chemotactic protein 2	GCP-2	Y08770
C-CHEMOKINES	LYMPHOTACTIN	SCM-1	D63789 D63790
CX_3C-CHEMOKINES	Fractalkine/neurotactin		U91835 U84487

The present invention also relates to the use of fragments, analogues and derivatives of the foregoing chemokines, as well as truncation isoforms of such chemokines which are known in the art.

The present invention also relates to therapeutic compositions comprising one or more chemokines, nucleic acids encoding one or more chemokines, derivatives, analogues, and/or truncation isoforms thereof, and nucleic acids encoding the same, that are effective to enhance the immune response of a subject to a vaccine.

In another preferred embodiment of the invention, nucleic acids comprising

nucleotide sequences encoding one or more chemokines or fragments or derivatives, including truncation isoforms, thereof, and encoding one or more antigens against which an immune response is desired, which coding sequences are operatively linked to gene regulatory sequences capable of directing the expression of the one or more chemokines and the one or more antigens upon introduction into a suitable cell, for example, but not limited to, the cell (of a subject), are administered to a subject such that the one or more chemokines, or fragments or derivatives, including truncation isoforms, thereof, and one or more antigens, are expressed in the subject.

For clarity of disclosure, and not by way of limitation, the detailed description of the invention is divided into the subsections which follow.

5.1. METHODS AND COMPOSITIONS TO ENHANCE THE EFFICACY OF A VACCINE

The present invention provides methods for enhancing the efficacy of a vaccine in a subject, which methods comprise administering to a subject an immunogenic amount of one or more purified antigens against which an immune response is desired in the subject in conjunction with an amount of one or more purified chemokines, or fragments, derivatives, analogues and/or truncation isoforms thereof, effective to enhance the immune response against the antigen. In one aspect, the purified chemokine(s), or fragment(s), derivative(s), analogue(s) and/or truncation isoforms thereof, are administered to the subject concurrently with (e.g., in the same composition with) the purified antigen or antigens against which an immune response is desired. In another, aspect, the purified chemokine(s), or fragment(s), derivative(s), analogue(s) and/or truncation isoforms thereof, are administered either before or after the administration of one or more purified antigens against which immunity is desired in the subject, but is administered within such time that the chemokine(s) enhance the immune response to the one or more antigens. For example, but not by way of limitation, the purified chemokine(s) are administered during the time that the subject mounts an immune response against the administered one or more antigens, or, the purified MDC is administered within, for example, but not limited to, 30 minutes, 1 hour, 5 hours, 10 hours, 1 day, 2 days of (preferably, after) administration of the one or more purified antigens against which immunity is desired.

In a preferred embodiment, the present invention provides compositions comprising an immunogenic amount of one or more purified antigens and an amount of purified MDC, or one or more fragments, derivatives or analogues thereof, effective to enhance the immune response to said antigen and, preferably, the composition further comprises a pharmaceutically acceptable carrier.

A preferred chemokine for use in the methods and compositions of the present invention is any MDC protein, fragment or derivative thereof, that is capable of enhancing the efficacy of a vaccine (for example, but not limited to, as determined by the assays described in Section 5.4, infra). In one specific embodiment, the MDC is purified full length MDC, preferably full length MDC having the amino acid sequence of SEQ ID NO: 2 (Figure 1B). In another embodiment, the MDC is a purified protein, the amino acid sequence of which consists of amino acid numbers 2-69 of SEQ ID NO: 2 (Figure 1B). In another specific embodiment, the MDC is a purified protein, the amino acid sequence of which consists of amino acid numbers 3-69 of SEQ ID NO: 2 (Figure 1B). In still another specific embodiment, the MDC is a purified protein, the N-terminal amino acid sequence of which consists of the amino acid sequence Tyr-Gly-Ala-Asn-Met-Glu-Asp-Ser-Val-Cys-Cys-Arg-Asp-Tyr-Val-Arg-Tyr-Arg-Leu (portion of SEQ ID NO: 2). In yet another specific embodiment, the MDC is a purified protein, the N-terminal amino acid sequence of which consists of the amino acid sequence Pro-Tyr-Gly-Ala-Asn-Met-Glu-Asp-Ser-Val-Cys-Cys-Arg (portion of SEQ ID NO: 2). In yet another specific embodiment, the MDC is a purified derivative of a protein, the N-terminal amino acid sequence of which protein consists of the amino acid sequence Tyr-Gly-Ala-Asn-Met-Glu-Asp-Ser-Val-Cys-Cys-Arg-Asp-Tyr-Val-Arg-Tyr-Arg-Leu (SEQ ID NO:2), which derivative has activity to enhance the efficacy of the vaccine. In yet another specific embodiment, the MDC is a purified derivative of a protein, the N-terminal amino acid sequence of which protein consists of the amino acid sequence Pro-Tyr-Gly-Ala-Asn-Met-Glu-Asp-Ser-Val-Cys-Cys-Arg (SEQ ID NO:2), which derivative has activity to enhance the efficacy of the vaccine.

In yet another specific embodiment, the chemokine is a purified derivative of the protein, which derivative has one or more insertions of or substitutions with one or more non-classical amino acids relative to a corresponding wildtype chemokine, which derivative will enhance the efficacy of the vaccine. In yet another specific

embodiment, the chemokine is a purified derivative of the protein that has only one or more conservative substitutions in sequence relative a corresponding wildtype chemokine, which derivative will enhance the efficacy of the vaccine. The chemokines useful in the present invention may be derived from any suitable source and obtained by any method known in the art, for example but not limited to the methods described in Section 5.2 infra.

Preferably, the chemokine(s) are of the same species as the subject to which the vaccine is administered. In a preferred embodiment, one or more human chemokines are administered to a human subject, e.g., human MDC is administered to a human subject, alone or in combination with another chemokine.

The present invention also provides a method to enhance the efficacy of a vaccine in a subject, which method comprises administering to a subject a purified first nucleic acid comprising a nucleotide sequence encoding an antigen against which an immune response is desired in a subject and a purified second nucleic acid comprising a nucleotide sequence encoding one or more chemokines, or fragment(s), derivative(s) or analogue(s) thereof, where the expression of the encoded antigen(s) and chemokine(s), or fragment(s), derivative(s) or analogue(s) thereof, are under control of one or more appropriate gene regulatory elements (which regulatory elements can be any regulatory element known in the art, for example, but not limited to, those regulatory elements described in Section 5.2 supra), such that, upon introduction of said first and second nucleic acids into a suitable cell (e.g., a cell of the subject), the antigen and chemokine(s), or fragment(s), derivative(s) or analogue(s) thereof, are coordinately expressed, *i.e.*, are expressed either at the same time or within an appropriate time period (*i.e.*, sufficient for the chemokine(s) to enhance the immune response against the antigen relative to a corresponding immune response in the absence of the chemokine) and the antigen(s) are expressed in an immunogenic amount and the chemokine(s), or fragment(s), derivative(s) or analogue(s) thereof, are expressed in an amount sufficient to enhance the immune response against the antigen(s). In a specific embodiment, the nucleotide sequences encoding the chemokine(s) and the antigen are present on separate nucleic acids. In another embodiment, the nucleotide sequences encoding the chemokine(s) and the antigen(s) are present on the same nucleic acid.

The present invention also provides compositions to enhance the

efficacy of a vaccine in a subject, which compositions comprise a purified first nucleic acid comprising a nucleotide sequence encoding one or more antigen(s) and a purified second nucleic acid comprising a nucleotide sequence encoding one or more chemokines, or fragments or derivatives, including truncation isoforms, thereof, wherein the nucleotide sequences encoding the antigens and the chemokine(s) are operably linked to one or more gene regulatory elements such that, upon introduction of said first and second nucleic acids into a suitable cell (e.g., a cell of the subject), the antigen(s) and chemokine(s) are expressed in a coordinated manner and the antigen(s) are expressed in an immunogenic amount and the chemokine(s) are expressed in an amount effective to enhance the immune response against the antigen, relative to a corresponding immune response in the absence of such chemokine(s).

The present invention also provides compositions to enhance the efficacy of a vaccine in a subject, which compositions comprise a purified first set of one or more purified nucleic acids comprising one or more nucleotide sequences encoding one or more antigens and a purified second set of one or more purified nucleic acids comprising a nucleotide sequence encoding one or more chemokines, or fragments, analogues, derivatives, (including truncation isoforms) thereof, wherein the nucleotide sequence(s) encoding the antigen(s) and the chemokine(s) are operably linked to one or more gene regulatory elements such that, upon introduction of said first and second sets of nucleic acids into a suitable cell (e.g., a cell of the subject), the antigen(s) and chemokine(s) are expressed in a coordinated manner and the antigen(s) are expressed in an immunogenic amount and the chemokine(s) are expressed in an amount effective to enhance the immune response against the antigen, relative to a corresponding immune response in the absence of such chemokine(s).

The present invention also provides compositions to enhance the efficacy of a vaccine in a subject, which compositions comprise a purified nucleic acid comprising a first set of one or more nucleotide sequences encoding one or more antigens and a second set of one ore more nucleotide sequence encoding one or more chemokines, or fragments, derivatives, or analogues thereof (including truncation isoforms), wherein the first and second sets of nucleotide sequences are operably linked to one or more gene regulatory elements such that, upon introduction into a suitable cell, the antigen(s) and the chemokine(s) are expressed in a coordinated manner and the antigen(s) are expressed in an immunogenic amount and the chemokine(s) are

expressed in an amount effective to enhance the immune response against the antigen(s).

Any nucleic acid comprising a nucleotide sequence encoding one or more chemokine proteins, or fragments or derivatives, thereof (including truncation isoforms), that are capable of enhancing the immune response to the antigen (for example, but not limited to, as determined by any of the assays described in Section 5.2., *infra*) can be used in the methods and compositions of the present invention.

In a preferred embodiment, the nucleotide sequence encodes MDC. In another embodiment, the MDC-encoding nucleotide consists of the nucleotide sequence of SEQ ID NO:1 (Figure 1A). In another specific embodiment, the method or composition of the invention uses a nucleic acid encoding an MDC derivative having deletional, insertional or substitutional mutations and combination thereof, which derivative has activity to enhance the immune response against an antigen in a subject.

Such compositions of nucleic acids encoding an antigen are often referred to as DNA vaccines.

Such DNA vaccines are produced by any method known in the art for constructing an expression plasmid vector containing the nucleotide sequences of the antigen(s) and/or chemokine(s) to be expressed which vector is suitable for expression of the encoded proteins in the subject or in cells recombinant for the expression vector, which cells are to be provided to the subject. Such expression vectors may contain various promoters, terminators and polyadenylation coding regions to control the expression of the encoded protein.

The DNA vaccine can be administered by any method known in the art for administration of DNA. The DNA vaccine may be delivered either directly, in which case the subject is directly exposed to the DNA vaccine such that the DNA enters and is expressed in cells of the subject, or indirectly, in which case, the DNA vaccine is first introduced into suitable cells by any method known in the art *in vitro*, then the cells containing the DNA vaccine are transplanted into the subject.

In a specific embodiment, the DNA vaccine is directly administered *in vivo*, where it is expressed to produce the encoded antigens and chemokine(s). This can be accomplished by any of numerous methods known in the art, e.g., by constructing it as part of an appropriate nucleic acid expression vector and administering it so that it becomes intracellular, *e.g.*, by infection using a defective or attenuated retroviral or

other viral vector (see U.S. Patent No. 4,980,286), or by direct injection of naked DNA, or by use of microparticle bombardment (*e.g.*, a gene gun; Biolistic, Dupont), or coating with lipids or cell-surface receptors or transfecting agents, encapsulation in liposomes, microparticles, or microcapsules, or by administering it in linkage to a peptide which is known to enter the nucleus, by administering it in linkage to a ligand subject to receptor-mediated endocytosis (see e.g., Wu and Wu, *J. Biol. Chem.* 262:4429-4432 (1987)) (which can be used to target cell types specifically expressing the receptors), etc. In another embodiment, a nucleic acid-ligand complex can be formed in which the ligand comprises a fusogenic viral peptide to disrupt endosomes, allowing the nucleic acid to avoid lysosomal degradation. In a preferred embodiment, the nucleic acid of a DNA vaccine is injected into the muscle of the subject to be immunized.

Another approach is to introduce the nucleic acid of the DNA vaccine into a cell prior to administration *in vivo* of the resulting recombinant cell. Such introduction can be carried out by any method known in the art, including but not limited to transfection, electroporation, microinjection, infection with a viral or bacteriophage vector containing the nucleic acid sequences, cell fusion, chromosome-mediated gene transfer, microcell-mediated gene transfer, spheroplast fusion, etc. Numerous techniques are known in the art for the introduction of foreign nucleic acid into cells (see *e.g.*, Loeffler and Behr, *Meth. Enzymol.* 217:599-618 (1993); Cohen et al., *Meth. Enzymol.* 217:618-644 (1993); Cline, *Pharmac. Ther.* 29:69-92 (1985)) and may be used in accordance with the present invention. Usually, the method of transfer includes the transfer of a selectable marker to the cells. The cells are then placed under selection to isolate those cells that have taken up and are expressing the transferred gene.

Cells into which a DNA vaccine can be introduced for purposes of immunization encompass any desired, available cell type, and include but are not limited to epithelial cells, endothelial cells, keratinocytes, fibroblasts, muscle cells, hepatocytes; blood cells such as T lymphocytes, B lymphocytes, monocytes, macrophages, neutrophils, eosinophils, megakaryocytes, granulocytes; various stem or progenitor cells, in particular hematopoietic stem or progenitor cells, *e.g.*, as obtained from bone marrow, umbilical cord blood, peripheral blood, fetal liver, etc.

The resulting recombinant cells can be delivered to a subject by various

methods known in the art. In a preferred embodiment, the recombinant cells are injected, *e.g.*, subcutaneously. In another embodiment, recombinant skin cells may be applied as a skin graft onto the patient. Recombinant blood cells (*e.g.*, hematopoietic stem or progenitor cells) are preferably administered intravenously. The cells can also be encapsulated in a suitable vehicle and then implanted in the subject (*see, e.g.*, Dionne et al. PCT Publication WO 92/19195, dated November 12, 1992). The amount of cells envisioned for use depends on the desired effect, subject state, etc., and can be determined by one skilled in the art.

By way of example, and not by way of limitation a DNA vaccine may be generated as described by Lekutis et al. for an HIV DNA vaccine (1997, *J. Immunol.* 158:4471-4477). Briefly, an expression vector is constructed with the promoter, enhancer and intron A of human cytomegalovirus (CMV) and the termination and polyadenylation sequences of bovine growth hormone in a plasmid backbone. Additionally, the nucleotide sequence for signal sequence of tissue plasminogen activator is either substituted for the signal sequence of the antigen, if the antigen has a signal sequence or is added onto the amino-terminus of the antigen, thereby eliminating the dependence on viral proteins for expression (*e.g.*, in the case of gp120 expression, rev and env proteins are required unless the HIV-1 signal sequence is so substituted). The resulting formulation is then injected intra-muscularly.

Further examples of DNA vaccines are set forth in Boyer et al. (1996, *J. Med. Primatol.*, 25:242-250), which describes the construction of a plasmid encoding the HIV-1 gp160 envelope glycoprotein as well as the *rev-tax* region cloned into pMAMneoBlue vector (Clonetech, Inc., Palo Alto, CA), and a vector encoding the envelope glycoprotein and rev from HIV-1 strain MN under the control of the CMV promoter. Another vector which can be used in the present invention is as described in Boyer et al. (1997, *Nature Medicine* 3:526-532) and contains expression cassettes encoding the envelope and Rev proteins of HIV-1 strain MN, and encoding the Gag/Pol proteins of HIV-1 strain IIIB.

For the practice of the present invention, the nucleotide sequence for the one or more chemokines, or fragments, derivatives, or analogues thereof, can either be incorporated into the same expression vector containing the nucleotide sequence encoding the antigen in such a manner that the chemokine(s) are expressed. Alternatively, the nucleotide sequence encoding the chemokine(s), or fragment(s),

derivative(s) or analogue(s) thereof, can be cloned into a separate expression vector (e.g., as described above for the expression vector containing the sequences coding for antigen) and the expression vector that expresses the antigen(s) mixed with the expression vector that expresses the chemokine(s). The mixture of the two expression vectors can then be administered to the subject.

The methods and compositions of the present invention may be used as a vaccine in a subject in which immunity for the antigen(s) is desired. Such antigens can be any antigen known in the art to be useful in a vaccine formulation. The methods and compositions of the present invention can be used to enhance the efficacy of any vaccine known in the art. The vaccine of the present invention may be used to enhance an immune response to infectious agents and diseased or abnormal cells, such as but not limited to bacteria, parasites, fungi, viruses, tumors and cancers. The compositions of the invention may be used to either treat or prevent a disease or disorder amenable to treatment or prevention by generating an immune response to the antigen provided in the composition. In one preferred embodiment, the antigen(s) are proteins, fragments or derivatives, including truncation isoforms, thereof, encoded by any genes of the HIV genome including the *env, gag, pol, nef, vif, rev,* and tat genes. In a more preferred embodiment, the antigen is an HIV-associated gp120 protein.

The methods and compositions of the present invention may be used to elicit a humoral and/or a cell-mediated response against the antigen(s) of the vaccine in a subject. In one specific embodiment, the methods and compositions elicit a humoral response against the administered antigen in a subject. In another specific embodiment, the methods and compositions elicit a cell-mediated response against the administered antigen in a subject. In a preferred embodiment, the methods and compositions elicit both a humoral and a cell-mediated response.

The subjects to which the present invention is applicable may be any mammalian or vertebrate species, which include, but are not limited to, cows, horses, sheep, pigs, fowl (*e.g.*, chickens), goats, cats, dogs, hamsters, mice and rats, monkeys, rabbits, chimpanzees, and humans. In a preferred embodiment, the subject is a human. The compositions and methods of the invention can be used to either prevent a disease or disorder, or to treat a particular disease or disorder, where an immune response against a particular antigen or antigens is effective to treat or prevent the

disease or disorder. Such diseases and disorders include, but are not limited to, viral infections, such as HIV, CMV, hepatitis, herpes virus, measles, etc, bacterial infections, fungal and parasitic infections, cancers, and any other disease or disorder amenable to treatment or prevention by eliciting an immune response against a particular antigen or antigens. In another preferred embodiment, the subject is infected or at risk of being infected with HIV virus.

In another preferred embodiment the invention provides methods and compositions to enhance the efficacy of an HIV vaccine, such a vaccine can be administered to either prevent or treat HIV.

5.2. CHEMOKINE GENES AND PROTEINS

Chemokine proteins and nucleic acids can be obtained by any method known in the art. Chemokine nucleotide and amino acid sequences are available in public databases such as Genbank and are also published in various references known to those of skill in the art. The gene bank accession numbers for the preferred chemokines of the present invention are provided in Table I, in Section 5 above. The ensuing discussion uses MDC by way of example, but applies equally to other chemokines as well.

The MDC nucleotide and amino acid sequences for, *inter alia*, human, are available in the public databases (e.g. Genbank accession No. U83171) also published in Godiska et al., 1997, *J. Exp. Med.* 185:1595-1604. The nucleotide sequence and the amino acid sequence for the human MDC are provided in Figures 1A and B (SEQ ID NOS:1 and 2, respectively).

Chemokines used herein include, but are not limited to, chemokines from mice, hamsters, dogs, cats, monkeys, rabbits, chimpanzees, and human. In one preferred embodiment, the chemokine is of human origin.

Any vertebrate cell potentially can serve as the nucleic acid source for the isolation of chemokine nucleic acids. The nucleic acid sequences encoding the chemokine(s) can be isolated from vertebrate, mammalian, human, porcine, bovine, feline, avian, equine, canine, as well as additional primate sources, etc. The DNA may be obtained by standard procedures known in the art from cloned DNA (e.g., a

DNA "library"), by chemical synthesis, by cDNA cloning, or by the cloning of genomic DNA, or fragments thereof, purified from the desired cell (see, for example, Sambrook et al., 1989, Molecular Cloning, A Laboratory Manual, 2d Ed., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York; Glover, D.M. (ed.), 1985, DNA Cloning: A Practical Approach, MRL Press, Ltd., Oxford, U.K. Vol. I, II.) Clones derived from genomic DNA may contain regulatory and intron DNA regions in addition to coding regions; clones derived from cDNA will contain only exon sequences. Whatever the source, the gene should be molecularly cloned into a suitable vector for propagation of the gene.

In the molecular cloning of the gene from cDNA, cDNA is generated from totally cellular RNA or mRNA by methods that are well known in the art. The gene may also be obtained from genomic DNA, where DNA fragments are generated (e.g. using restriction enzymes or by mechanical shearing), some of which will encode the desired gene. The linear DNA fragments can then be separated according to size by standard techniques, including but not limited to, agarose and polyacrylamide gel electrophoresis and column chromatography.

Once the DNA fragments are generated, identification of the specific DNA fragment containing all or a portion of the chemokine gene may be accomplished in a number of ways.

A preferred method for isolating a chemokine gene is by the polymerase chain reaction (PCR), which can be used to amplify the desired chemokine sequence in a genomic or cDNA library or from genomic DNA or cDNA that has not been incorporated into a library. Oligonucleotide primers which would hybridize to chemokine sequences can be used as primers in PCR.

Additionally, a portion of the chemokine (of any species) gene or its specific RNA, or a fragment thereof, can be purified (or an oligonucleotide synthesized) and labeled, the generated DNA fragments may be screened by nucleic acid hybridization to the labeled probe (Benton, W. and Davis, R., 1977, Science 196:180; Grunstein, M. And Hogness, D., 1975, Proc. Natl. Acad. Sci. U.S.A. 72:3961). Those DNA fragments with substantial homology to the probe will hybridize. Chemokine nucleic acids can be also identified and isolated by expression cloning using, for example, anti-chemokine antibodies for selection.

Alternatives to obtaining the chemokine DNA by cloning or amplification

include, but are not limited to, chemically synthesizing the gene sequence itself from the known chemokine sequence or making cDNA to the mRNA which encodes the chemokine protein. Other methods are possible and within the scope of the invention. Once a clone has been obtained, its identity can be confirmed by nucleic acid sequencing (by any method well known in the art) and comparison to known chemokine sequences. DNA sequence analysis can be performed by any techniques known in the art, including but not limited to the method of Maxam and Gilbert (1980, Meth. Enzymol. 65:499-560), the Sanger dideoxy method (Sanger, F., et al., 1977, Proc. Natl. Acad. Sci. U.S.A. 74:5463), the use of T7 DNA polymerase (Tabor and Richardson, U.S. Patent No. 4,795,699), use of an automated DNA sequenator (e.g., Applied Biosystems, Foster City, CA) or the method described in PCT Publication WO 97/ 15690.

Nucleic acids which are hybridizable to a chemokine nucleic acid, or to a nucleic acid encoding a chemokine derivative can be isolated, by nucleic acid hybridization under conditions of low, high, or moderate stringency (see also Shilo and Weinberg, 1981, Proc. Natl. Acad. Sci. USA 78:6789-6792). For example, the nucleic acid of SEQ ID No: 1 is hybridizable to an MDC nucleic acid.

Chemokine proteins and derivatives, analogs and fragments of chemokine proteins can be obtained by any method known in the art, including but not limited to recombinant expression methods, purification from natural sources, and chemical synthesis.

For example, chemokines can be obtained by recombinant protein expression techniques. For recombinant expression, the chemokine gene or portion thereof is inserted into an appropriate cloning vector for expression in a particular host cell. A large number of vector-host systems known in the art may be used. Possible vectors include, but are not limited to, plasmids or modified viruses, but the vector system must be compatible with the host cell used. Such vectors include, but are not limited to, bacteriophages such as lambda derivatives, or plasmids such as pBR322 or pUC plasmid derivatives or the Bluescript vector (Stratagene). The insertion into a cloning vector can, for example, be accomplished by ligating the DNA fragment into a cloning vector which has complementary cohesive termini. However, if the complementary restriction sites used to fragment the DNA are not present in the cloning vector, the ends of the DNA molecules may be enzymatically modified. Alternatively, any site

desired may be produced by ligating nucleotide sequences (linkers) onto the DNA termini; these ligated linkers may comprise specific chemically synthesized oligonucleotides encoding restriction endonuclease recognition sequences. In an alternative method, the cleaved vector and chemokine gene may be modified by homopolymeric tailing. Recombinant molecules can be introduced into host cells via transformation, transfection, infection, electroporation, etc., so that many copies of the gene sequence are generated.

In an alternative method, the desired gene may be identified and isolated after insertion into a suitable cloning vector in a "shot gun" approach. Enrichment for the desired gene, for example, by size fractionation, can be done before insertion into the cloning vector.

In specific embodiments, transformation of host cells with recombinant DNA molecules that incorporate the isolated chemokine gene, cDNA, or synthesized DNA sequence enables generation of multiple copies of the gene. Thus, the gene may be obtained in large quantities by growing transformants, isolating the recombinant DNA molecules from the transformants and, when necessary, retrieving the inserted gene from the isolated recombinant DNA.

The nucleotide sequence coding for a chemokine protein or a functionally active analog or fragment or other derivative thereof, can be inserted into an appropriate expression vector, *i.e.*, a vector which contains the necessary elements for the transcription and translation of the inserted protein-coding sequence. The necessary transcriptional and translational signals can also be supplied by the native chemokine gene and/or its flanking regions. A variety of host-vector systems may be utilized to express the protein-coding sequence. These include but are not limited to mammalian cell systems infected with virus (e.g., vaccinia virus, adenovirus, etc.); insect cell systems infected with virus (*e.g.*, baculovirus); microorganisms such as yeast containing yeast vectors, or bacteria transformed with bacteriophage, DNA, plasmid DNA, or cosmid DNA. The expression elements of vectors vary in their strengths and specificities. Depending on the host-vector system utilized, any one of a number of suitable transcription and translation elements may be used.

Any of the methods previously described for the insertion of DNA fragments into a vector may be used to construct expression vectors containing a chimeric gene consisting of appropriate transcriptional/translational control signals and the protein

coding sequences. These methods may include *in vitro* recombinant DNA and synthetic techniques and *in vivo* recombinants (genetic recombination). Expression of nucleic acid sequence encoding a chemokine protein or peptide fragment may be regulated by a second nucleic acid sequence so that the chemokine protein or peptide is expressed in a host transformed with the recombinant DNA molecule. For example, expression of a chemokine protein may be controlled by any promoter/enhancer element known in the art. Promoters which may be used to control chemokine expression include, but are not limited to, the SV40 early promoter region (Bernoist and Chambon, 1981, Nature 290:304-310), the promoter contained in the 3' long terminal repeat of Rous sarcoma virus (Yamamoto, et al., 1980, Cell 22:787-797), the herpes thymidine kinase promoter (Wagner et al., 1981, Proc. Natl. Acad. Sci. U.S.A. 78:1441-1445), the regulatory sequences of the metallothionein gene (Brinster et al., 1982, Nature 296:39-42); prokaryotic expression vectors such as the β-lactamase promoter (Villa-Kamaroff, et al., 1978, Proc. Natl. Acad. Sci. U.S.A. 75:3727-3731), or the tac promoter (DeBoer, et al., 1983, Proc. Natl. Acad. Sci. U.S.A. 80:21-25); see also "Useful proteins from recombinant bacteria" in Scientific American, 1980, 242:74-94; promoter elements from yeast or other fungi such as the Gal 4 promoter, the ADC (alcohol dehydrogenase) promoter, PGK (phosphoglycerol kinase) promoter, alkaline phosphatase promoter, and the following animal transcriptional control regions, which exhibit tissue specificity and have been utilized in transgenic animals: elastase I gene control region which is active in pancreatic acinar cells (Swift et al., 1984, Cell 38:639-646; Ornitz et al., 1986, Cold Spring Harbor Symp. Quant. Biol. 50:399-409; MacDonald, 1987, Hepatology 7:425-515); insulin gene control region which is active in pancreatic beta cells (Hanahan, 1985, Nature 315:115-122), immunoglobulin gene control region which is active in lymphoid cells (Grosschedl et al., 1984, Cell 38:647-658; Adames et al., 1985, Nature 318:533-538; Alexander et al., 1987, Mol. Cell. Biol. 7:1436-1444), mouse mammary tumor virus control region which is active in testicular, breast, lymphoid and mast cells (Leder et al., 1986, Cell 45:485-495), albumin gene control region which is active in liver (Pinkert et al., 1987, Genes and Devel. 1:268-276), alpha-fetoprotein gene control region which is active in liver (Krumlauf et al., 1985, Mol. Cell. Biol. 5:1639-1648; Hammer et al., 1987, Science 235:53-58; alpha 1-antitrypsin gene control region which is active in the liver (Kelsey et al., 1987, Genes and Devel. 1:161-171), beta-globin gene control region

which is active in myeloid cells (Mogram et al., 1985, Nature 315:338-340; Kollias et al., 1986, Cell 46:89-94), myelin basic protein gene control region which is active in oligodendrocyte cells in the brain (Readhead et al., 1987, Cell 48:703-712), myosin light chain-2 gene control region which is active in skeletal muscle (Sani, 1985, Nature 314:283-286), and gonadotropic releasing hormone gene control region which is active in the hypothalamus (Mason et al., 1986, Science 234:1372-1378).

For example, a vector can be used that comprises a promoter operably linked to an chemokine-encoding nucleic acid, one or more origins of replication, and, optionally, one or more selectable markers (*e.g.*, an antibiotic resistance gene).

In a specific embodiment, an expression construct is made by subcloning a chemokine coding sequence into the *Eco*RI restriction site of each of the three pGEX vectors (Glutathione S-Transferase expression vectors; Smith and Johnson, 1988, Gene 7:31-40). This allows for the expression of the chemokine protein product from the subclone in the correct reading frame.

Expression vectors containing chemokine gene inserts can be identified by three general approaches: (a) nucleic acid hybridization, (b) presence or absence of "marker" gene functions, and (c) expression of inserted sequences. In the first approach, the presence of a chemokine gene inserted in an expression vector can be detected by nucleic acid hybridization using probes comprising sequences that are homologous to an inserted chemokine gene. In the second approach, the recombinant vector/host system can be identified and selected based upon the presence or absence of certain "marker" gene functions (*e.g.*, thymidine kinase activity, resistance to antibiotics, transformation phenotype, occlusion body formation in baculovirus, etc.) caused by the insertion of a chemokine gene in the vector. For example, if the chemokine gene is inserted within the marker gene sequence of the vector, recombinants containing the chemokine insert can be identified by the absence of the marker gene function. In the third approach, recombinant expression vectors can be identified by assaying the product expressed by the recombinant. Such assays can be based, for example, on the physical or functional properties of the chemokine protein in *in vitro* assay systems, *e.g.*, binding with anti-chemokine antibody or the chemokine's receptor.

Once a particular recombinant DNA molecule is identified and isolated, several methods known in the art may be used to propagate it. Once a suitable host

system and growth conditions are established, recombinant expression vectors can be propagated and prepared in quantity. As previously explained, the expression vectors which can be used include, but are not limited to, the following vectors or their derivatives: human or animal viruses such as vaccinia virus or adenovirus; insect viruses such as baculovirus; yeast vectors; bacteriophage vectors (*e.g.*, lambda), and plasmid and cosmid DNA vectors, to name but a few.

In addition, a host cell strain may be chosen which modulates the expression of the inserted sequences, or modifies and processes the gene product in the specific fashion desired. Expression from certain promoters can be elevated in the presence of certain inducers; thus, expression of the genetically engineered protein may be controlled. Furthermore, different host cells have characteristic and specific mechanisms for the translational and post-translational processing and modification (*e.g.*, glycosylation, phosphorylation of proteins. Appropriate cell lines or host systems can be chosen to ensure the desired modification and processing of the foreign protein expressed. For example, expression in a bacterial system can be used to produce an unglycosylated core protein product. Expression in yeast will produce a glycosylated product. Expression in mammalian cells can be used to ensure "native" glycosylation of a heterologous protein. Furthermore, different vector/host expression systems may effect processing reactions to different extents.

In other specific embodiments, the chemokine protein(s), fragment(s), analogue(s), or derivative(s) may be expressed as a fusion, or chimeric protein product (comprising the protein, fragment, analog, or derivative joined via a peptide bond to a heterologous protein sequence (of a different protein)). Such a chimeric product can be made by ligating the appropriate nucleic acid sequences encoding the desired amino acid sequences to each other by methods known in the art, in the proper coding frame, and expressing the chimeric product by methods commonly known in the art. Alternatively, such a chimeric product may be made by protein synthetic techniques, *e.g.*, by use of a peptide synthesizer. In a specific embodiment, a chimeric protein containing all or a portion of the chemokine is joined via a peptide bond to all or a portion of an antigen against which immunity is desired.

Both cDNA and genomic sequences can be cloned and expressed.

The chemokine protein(s) may also be isolated and purified by standard methods including chromatography (*e.g.*, ion exchange, affinity, and sizing column

chromatography), centrifugation, differential solubility, or by any other standard technique for the purification of proteins. The functional properties may be evaluated using any suitable assay (see Section 5.5). Alternatively, the protein can be synthesized by standard chemical methods known in the art (e.g., see Hunkapiller, M., et al., 1984, Nature 310:105-111). The chemokine-encoding nucleic acid sequence(s) can be mutated *in vitro* or *in vivo*, to create and/or destroy translation, initiation, and/or termination sequences, or to create variations in coding regions. Any technique for mutagenesis known in the art can be used, including, but not limited to, *in vitro* site-directed mutagenesis (Hutchinson et al., 1978, *J. Biol. Chem* 253:6551), use of TAB linkers (Pharmacia), mutation-containing PCR primers, etc.

The experimentation involved in mutagenesis consists primarily of site-directed mutagenesis followed by phenotypic testing of the altered gene product. Some of the more commonly employed site-directed mutagenesis protocols take advantage of vectors that can provide single stranded as well as double stranded DNA, as needed. Generally, the mutagenesis protocol with such vectors is as follows. A mutagenic primer, *i.e.*, a primer complementary to the sequence to be changed, but consisting of one or a small number of altered, added, or deleted bases, is synthesized. The primer is extended *in vitro* by a DNA polymerase and, after some additional manipulations, the now double-stranded DNA is transfected into bacterial cells. Next, by a variety of methods, the desired mutated DNA is identified, and the desired protein is purified from clones containing the mutated sequence. For longer sequences, additional cloning steps are often required because long inserts (longer than 2 kilobases) are unstable in those vectors. Protocols are known to these skilled in the art and kits for site-directed mutagenesis are widely available from biotechnology supply companies, for example from Amersham Life Science, Inc. (Arlington Heights, IL) and Stratagene Cloning Systems (La Jolla, CA).

In other specific embodiments, the chemokine derivative(s) or analogue(s) may be expressed as a fusion, or chimeric protein product (comprising the protein, fragment, analogue, or derivative joined via a peptide bond to a heterologous protein sequence (of a different protein)). Such a chimeric product can be made by ligating the appropriate nucleic acid sequences encoding the desired amino acid sequences to each other by methods known in the art, in the proper coding frame, and expressing the chimeric product by methods commonly known in the art.

In addition, chemokine proteins, derivatives (including fragments and chimeric proteins), and analogues can be chemically synthesized. *See, e.g.*, Clark-Lewis et al., 1991, *Biochem.* 30:3128-3135 and Merrifield, 1963, *J. Amer. Chem. Soc.* 85:2149-2156. For example, chemokines, derivatives and analogues can be synthesized by solid phase techniques, cleaved from the resin, and purified by preparative high performance liquid chromatography (*e.g.*, *see* Creighton, 1983, Proteins, Structures and Molecular Principles, W.H. Freeman and Co., N.Y., pp. 50-60). Chemokines, derivatives and analogues that are proteins can also be synthesized by use of a peptide synthesizer. The composition of the synthetic peptides may be confirmed by amino acid analysis or sequencing (e.g., the Edman degradation procedure; *see* Creighton, 1983, Proteins, Structures and Molecular Principles, W.H. Freeman and Co., N.Y., pp. 34-49).

The chemokine proteins, derivatives, or analogues of the invention may be synthesized in their entirety by the sequential addition of amino acid residues or alternatively as fragment subcomponents which may be combined using techniques well known in the art, such as, for example, fragment condensation (Shin et al., 1992, *Biosci. Biotech. Biochem.* 56:404-408; Nyfeler et al., 1992, Peptides, Proc. 12th Amer. Pep. Soc., Smith and Rivier (eds), Leiden, pp 661-663); and Nokihara et al., 1990, Protein Research Foundation, Yanaihara (ed), Osaka, pp 315-320).

In a less preferred embodiment, chemokine derivatives can be obtained by proteolysis of the protein followed by purification using standard methods such as those described above (e.g., immunoaffinity purification).

In another alternate embodiment, native chemokine proteins can be purified from natural sources, by standard methods such as those described above (e.g., immunoaffinity purification).

5.3. COMPOSITION FORMULATIONS AND METHODS OF ADMINISTRATION

The composition formulations of the invention comprise an effective immunizing amount of an immunologically active ingredient, i.e., one or more antigens, and an amount of one or more chemokine(s), or fragment(s) or derivative thereof, effective to enhance the immune response against the antigen in a subject, and a pharmaceutically acceptable carrier or excipient. In a specific embodiment, the

chemokines are selected from the group consisting of Macrophage-derived chemokine, Monocyte chemotactic protein 1, Monocyte chemotactic protein 2, Monocyte chemotactic protein 3, Monocyte chemotactic protein 4, activated macrophage specific chemokine 1, Macrophage inflammatory protein 1 alpha, Macrophage inflammatory protein 1 beta, Macrophage inflammatory protein 1 gamma, Macrophage inflammatory protein 1 delta, Macrophage inflammatory protein 2 alpha, Macrophage inflammatory protein 3 alpha, Macrophage inflammatory protein 3 beta, Regulated upon activation, normal T cell expressed and secreted (and its variants), I-309, EBI1-ligand chemokine, Pulmonary and activation regulated chemokine, Liver and activation-regulated chemokine, Thymus and activation regulated chemokine, Eotaxin (and variants), Human CC chemokine 1, Human CC chemokine 2, Human CC chemokine 3, IL-10-inducible chemokine, liver-expressed chemokine, 6Ckine, Exodus 1, Exodus 2, Exodus 3, thymus-expressed chemokine, Secondary Lymphoid tissue chemokine, Lymphocyte and Monocyte chemoattractant; Monotactin, Activation-induced, chemokine-related molecule, Myeloid progenitor inhibitory factor-1, Myeloid progenitor inhibitory factor-2, Stromal cell-derived factor 1 alpha, Stromal cell-derived factor 1 beta, B-cell-attracting chemokine 1, HuMIG, H174, Interferon-stimulated T-cell alpha chemoattractant, Interleukin-8, IP-10, platelet factor 4, growth-regulated gene-alpha, growth-regulated gene-beta, growth-regulated gene-gamma, Neutrophil-activating protein 2, ENA-78, granulocyte chemotactic protein 2, LYMPHOTACTIN, and Fractalkine/neurotactin.

Pharmaceutically acceptable carriers or excipients are well known in the art and include but are not limited to saline, buffered saline, dextrose, water, glycerol, ethanol, sterile isotonic aqueous buffer, and combinations thereof. One example of such an acceptable carrier is a physiologically balanced culture medium containing one or more stabilizing agents such as stabilized, hydrolyzed proteins, lactose, etc. The carrier is preferably sterile. The formulation should suit the mode of administration.

In addition, if desired, the vaccine or composition preparation may also include minor amounts of auxiliary substances such as wetting or emulsifying agents, pH buffering agents, and/or adjuvants which enhance the effectiveness of the vaccine or composition. Suitable adjuvants may include, but are not limited to: mineral gels,

e.g., aluminum hydroxide; surface active substances such as lysolecithin, pluronic polyols; polyanions; peptides; oil emulsions; alum, MDP, N-acetyl-muramyl-L-threonyl-D-isoglutamine (thr-MDP), N-acetyl-nor-muramyl-L-alanyl-D-isoglutamine, and N-acetylmuramyl-L-alanyl-D-isoglutaminyl-L-alanine-2-(1'-2'-dipalmitoyl-sn-glycero-3-hydroxyphosphoryloxy)-ethylamine. The effectiveness of an adjuvant may be determined by comparing the induction of antibodies directed against a MDC-containing composition in the presence and in the absence of various adjuvants.

In instances where the recombinant antigen is a hapten, *i.e.*, a molecule that is antigenic in that it can react selectively with cognate antibodies, but not immunogenic in that it cannot elicit an immune response, the hapten may be covalently bound to a carrier or immunogenic molecule; for instance, a large protein such as serum albumin will confer immunogenicity to the hapten coupled to it. The hapten-carrier may be formulated for use as a vaccine.

The composition can be a liquid solution, suspension, emulsion, tablet, pill, capsule, sustained release formulation, or powder. Oral formulation can include standard carriers such as pharmaceutical grades of mannitol, lactose, starch, magnesium stearate, sodium saccharine, cellulose, magnesium carbonate, etc.

The chemokine(s), or fragment(s) or derivative(s) thereof, and/or the antigen(s) may be formulated into the composition as neutral or salt forms. Pharmaceutically acceptable salts include the acid addition salts (formed with free amino groups of the peptide) and which are formed with inorganic acids, such as, for example, hydrochloric or phosphoric acids, or organic acids such as acetic, oxalic, tartaric, maleic, and the like. Salts formed with free carboxyl groups may also be derived from inorganic bases, such as, for example, sodium potassium, ammonium, calcium, or ferric hydroxides, and such organic bases as isopropylamine, trimethylamine, 2-ethylamino ethanol, histidine, procaine and the like.

The vaccines of the invention may be multivalent or univalent. Multivalent vaccines are made from recombinant viruses that direct the expression of more than one antigen.

An effective dose (immunizing amount) is that amount sufficient to produce an immune response to the antigen(s) in the host to which the vaccine preparation is administered. The precise dose of the composition to be employed in the formulation will depend on the route of administration, and the nature of the subject to be

immunized, and should be decided by the practitioner according to standard clinical techniques. Effective doses of the vaccines or compositions of the present invention may also be extrapolated from dose-response curves derived from animal model test systems.

The invention also provides a pharmaceutical pack or kit comprising one or more containers comprising one or more of the ingredients of the composition formulations of the invention. Associated with such container(s) can be a notice in the form prescribed by a governmental agency regulating the manufacture, use or sale of pharmaceuticals or biological products, which notice reflects approval by the agency of manufacture, use or sale for human administration.

Generally, the ingredients are supplied either separately or mixed together in unit dosage form, for example, as a dry lyophilized powder or water free concentrate in a hermetically sealed container such as an ampoule or sachette indicating the quantity of active agent. Where the composition is administered by injection, an ampoule of sterile diluent can be provided so that the ingredients may be mixed prior to administration.

In a specific embodiment, a lyophilized immunologically active ingredient and one or more chemokine polypeptide(s) of the invention are provided in a first container; a second container comprises diluent consisting of an aqueous solution of 50% glycerin, 0.25% phenol, and an antiseptic (e.g., 0.005% brilliant green).

Many methods may be used to introduce the composition formulations of the invention; these include but are not limited to oral, intradermal, intramuscular, intraperitoneal, intravenous, subcutaneous, intranasal routes, and via scarification (scratching through the top layers of skin, e.g., using a bifurcated needle).

The DNA vaccines of the invention can be administered by any method known in the art for delivery of DNA to subject (for example, as described in Section 5.3 supra)

5.4. DETERMINATION OF COMPOSITION EFFICACY

The activity of one or more chemokines, or a fragment, derivative or analogue thereof, to enhance immune response to an antigen can be determined by monitoring the immune response in test animals following immunization with a composition containing the chemokine(s) and an antigen and comparing the response to that following immunization with the antigen in the absence of the chemokine(s). Generation of a humoral (antibody) response and/or cell-mediated immunity, may be taken as an indication of an immune response. Test animals may include mice, hamsters, dogs, cats, monkeys, rabbits, chimpanzees, etc., and eventually human subjects. Assays for humoral and cell-mediated immunity are well known in the art.

Methods of introducing the composition may include oral, intracerebral, intradermal, intramuscular, intraperitoneal, intravenous, subcutaneous, intranasal or any other standard routes of immunization. The immune response of the test subjects can be analyzed by various approaches well known in the art, such as but not limited to: testing the reactivity of the resultant immune serum to the antigen of the chemokine-containing vaccine, as assayed by known techniques, e.g., immunosorbant assay (ELISA), immunoblots, radioimmunoprecipitations, etc.

As one example of suitable animal testing, a composition of the present invention may be tested in mice for the ability to enhance an antibody response to an antigen (using for example, but not limited to, the method as described in Section 6, *infra*) and the delayed-type hypersensitivity (DTH) response (also described in Section 6 infra), measured by an increase in footpad swelling after inoculation in the footpad of the test animal, as compared to the measurements in animals administered the antigen in a composition not containing chemokine. For example, as test animals BALB/c mice may be used. The test group each receives an inoculation with fixed amount of antigen and varying amount of one or more chemokines. The control group receives an inoculation of comparable amount of antigen alone.

Serum samples may be drawn from the mice after the final inoculation (for example every one or two weeks after inoculation), and serum is analyzed for antibodies against the antigen using known methods in the art, e.g., using an ELISA. DTH responses to the antigen may be measured after the final inoculation (e.g. within 1-7 days). An increase in the serum titer of antibodies recognizing the antigen and/or

an increase in footpad swelling in the animals receiving the antigen-compositions containing the chemokine(s) as compared to the serum titer of antibodies against the antigen and/or the footpad swelling in the animals receiving the antigen composition not containing the chemokine(s), indicates that the chemokine(s) enhance the immune response to antigen. An increase in the serum titer of antibodies recognizing the antigen and/or an increase in footpad swelling in the animals receiving the antigen-compositions containing the chemokines as compared to the serum titer of antibodies against the antigen and/or the footpad swelling in the animals receiving the antigen composition not containing chemokine(s), indicates that the chemokine(s) enhances the immune response to antigen. An increase in the serum titer of antibodies recognizing the antigen and/or an increase in footpad swelling in the animals receiving the antigen-compositions containing MDC as compared to the serum titer of antibodies against the antigen and/or the footpad swelling in the animals receiving the antigen composition not containing MDC, indicates that the MDC enhances the immune response to antigen. An increase in the serum titer of antibodies recognizing the antigen and/or an increase in footpad swelling in the animals receiving the antigen-compositions containing MDC as compared to the serum titer of antibodies against the antigen and/or the footpad swelling in the animals receiving the antigen composition not containing MDC, indicates that the MDC enhances the immune response to antigen.

6. EXAMPLE: IMMUNIZATION WITH MDC-CONTAINING COMPOSITION

The following experiment illustrates the evaluation of whether MDC will act as an adjuvant for a protein antigen and enhance the efficacy of a vaccine. However, it will be appreciated that the description applies equally to other chemokines and combinations of chemokines.

6.1. MATERIALS AND METHODS

6.1.1. ANIMALS AND REAGENTS

BALB/c mice are purchased from Harlan-Sprague-Dawley (Indianapolis, IN).

Human MDC (hMDC) was obtained from CD8$^+$ T cell clones immortalized *in vitro* prepared as previously described (Markham et al., 1983 *Int. J. Cancer* 31:413; Markham et al. 1984, *Int. J. Cancer* 33:13). One such immortalized CD8$^+$ T cell clone, F3b Clone 19, was adapted to growth in serum-free medium by the following procedure and used for further studies. F3b Clone 19 cells were grown in complete medium containing rIL-2 (16 ng/ml) at 37?C in a CO_2 incubator. After expanding the culture to 200 ml, the cells were pelleted and resuspended in RPMI medium containing HB101 (Irvine Scientific) supplemented with 16 ng/ml of rIL-2, 1% glutamine and 1% penicillin/streptomycin. The cells were grown to full confluence and the medium harvested by centrifugation at 670 x g for 10 minutes.

Human MDC (hMDC) was purified from F3b Clone 19 as described in Pal et al., 1997, *Science* 278:695-698. Briefly, the cell free culture supernatant from F3b Clone 19 was clarified by high speed centrifugation and fractionated by heparin affinity chromatography, taking advantage of the heparin binding characteristics of chemokines (Witt and Lander, 1994, *Current Biology* 4:394; Proost et al., 1996, *Method: A Companion to Methods in Enzymology* 10:82). Culture supernatant (1200 ml) from F3b Clone 19, grown to high cell density in serum-free medium supplemented with rIL-2 was clarified by high speed centrifugation (100,000 x g for 60 minutes at 4?C) and applied to a 5 ml HiTrap heparin affinity FPLC column (Pharmacia) equilibrated in 10 mM Tris-HCl, pH 7.6 containing 0.1 M NaCl (column buffer). The column was then washed extensively with column buffer and the bound proteins eluted from the column with 10 mM Tris-HCl, pH 7.6 containing 2.0 M NaCl at a flow rate of 0.5 to 1 ml/minute. Virtually all of the HIV suppressive activity effective against primary NSI and SI isolates and HIV-1 $_{IIIB}$ was recovered in the column eluate (data not shown). The heparin affinity column eluate was brought to pH 2.0 by addition of trifluoracetic acid (TFA) and subjected to reversed phase HPLC on a PEEK C-18 column (Waters Instruments) equilibrated in H_2O containing 0.1 % TFA. Proteins bound to the column were eluted with a 5 minute linear gradient of aqueous acetonitrile (0 to 35 %) containing 0.1% TFA. After 10 minutes at 35% acetonitrile, the column was further developed with a 60 minute linear gradient of 35-70% aqueous acetonitrile in TFA. The flow rate was maintained at 0.5 to 1 ml/minute. The fractions obtained were then tested for suppressor activity in the acute infectivity assay using HIV-1$_{IIIB}$. Active fractions were pooled, diluted twofold in H_2O with 0.1 % TFA

and reapplied to the column. The column was then developed with a 30 minute linear aqueous acetonitrile gradient (0-60%) containing 0.1% TFA at a flow rate of 0.5 to 1 ml/minute. The fractions obtained were assayed as above. Active fractions were pooled, diluted with H_2O/0.1 % TFA and fractionated under the same conditions to obtain a single protein peak. The fraction corresponding to the peak and flanking fractions were tested in the infectivity assay to verify that suppressor activity was cofractionated with the protein.

Suppressive activity against HIV-1_{IIIB} in the absence of cytotoxic effects consistently copurified with a single protein peak that appeared as a homogeneous 8 kDa band when analyzed by SDS-polyacrylamide gel electrophoresis. This protein was not reactive in ELISAs for RANTES, MIP-1α or MIP-1β (R&D Systems).

Recombinant gp120 protein derived from HIV-1 IIIB isolate is purchased from Intracel (Foster City, CA).

6.1.2 IMMUNIZATION OF MICE

The hMDC and the gp120 is resuspended in a total volume of 50 µl of phosphate-buffered saline (PBS). Mice are divided into 5 groups with 3-4 mice in each group. Groups 1-4 are inoculated with 10 µg gp120 and 0.3 µg, 0.1 µg, 0.03 µg, and 0.01 µg of hMDC, respectively. As a control, group 5 is inoculated with 10 µg of gp120 in the absence of hMDC. For primary inoculation, each group of mice is inoculated with 10 µl of the hMDC and gp120 solution via footpad. Two to three weeks after the primary inoculation, each mouse is given the same does of hMDC/gp120 that is used in primary inoculation.

6.1.3 ELISA ASSAY

Serum samples are collected one week after the second inoculation via tail vein bleed. gp120 serum responses are measured using standard gp120 antibody ELISA assays.

6.1.4 DTH ASSAY

The delayed-type hypersensitivity (DTH) response is measured from 1-7 days after the second inoculation. A caliper is to be used to measure footpad swelling.

6.2. RESULTS

Mice inoculated with hMDC/gp120 are expected to have greater serum antibody and DTH responses than mice inoculated with gp120 alone. The improved responses will be reflected in either increased titers of serum antibody responses or increased footpad swelling. A dose response effect is expected - increasing the dose of hMDC used is expected to cause a corresponding improvement in the serum and DHT gp120-specific responses.

7. EXAMPLE: OTHER CHEMOKINES AND COMBINATIONS OF CHEMOKINES

The foregoing experiments can be repeated using other chemokines and combinations of chemokines. For example, the experiments are preferably repeated using one or more chemokines selected from the group consisting of: Macrophage-derived chemokine, Monocyte chemotactic protein 1, Monocyte chemotactic protein 2, Monocyte chemotactic protein 3, Monocyte chemotactic protein 4, activated macrophage specific chemokine 1, Macrophage inflammatory protein 1 alpha, Macrophage inflammatory protein 1 beta, Macrophage inflammatory protein 1 gamma, Macrophage inflammatory protein 1 delta, Macrophage inflammatory protein 2 alpha, Macrophage inflammatory protein 3 alpha, Macrophage inflammatory protein 3 beta, Regulated upon activation, normal T cell expressed and secreted (and its variants), I-309, EBI1-ligand chemokine, Pulmonary and activation regulated chemokine, Liver and activation-regulated chemokine, Thymus and activation regulated chemokine, Eotaxin (and variants), Human CC chemokine 1, Human CC chemokine 2, Human CC chemokine 3, IL-10-inducible chemokine, liver-expressed chemokine., 6Ckine, Exodus 1, Exodus 2, Exodus 3, thymus-expressed chemokine, Secondary Lymphoid tissue chemokine, Lymphocyte and Monocyte chemoattractant; Monotactin, Activation-induced, chemokine-related molecule, Myeloid progenitor inhibitory factor-1, Myeloid progenitor inhibitory factor-2, Stromal cell-derived factor 1 alpha, Stromal cell-derived factor 1 beta, B-cell-attracting chemokine 1, HuMIG, H174, Interferon-stimulated T-cell alpha chemoattractant, Interleukin-8, IP-10, platelet factor 4, growth-regulated gene-alpha, growth-regulated gene-beta, growth-regulated gene-gamma, Neutrophil-activating protein 2, ENA-78, granulocyte chemotactic protein 2, LYMPHOTACTIN, and Fractalkine/neurotactin.

The present invention is not to be limited in scope by the specific embodiments described herein. Indeed, various modifications of the invention in addition to those described herein will become apparent to those skilled in the art from the foregoing description. Such modifications are intended to fall within the scope of the appended claims.

Various references are cited herein, the disclosures of which are incorporated by reference in their entireties.

THE CLAIMS:

1. A method to enhance the efficacy of a vaccine in a subject comprising administering to the subject an immunogenic amount of one or more purified antigens against which an immune response is desired in the subject and an amount of one or more chemokines, or purified fragments or derivatives thereof, effective to enhance the efficacy of said vaccine.

2. The method of claim 1, wherein the one or more chemokines are selected from a chemokine class selected from the group consisting of: CC, CXC, C-C and CX3C.

3. The method of claim 1, wherein the one or more chemokines are selected from the group consisting of: Macrophage-derived chemokine, Monocyte chemotactic protein 1, Monocyte chemotactic protein 2, Monocyte chemotactic protein 3, Monocyte chemotactic protein 4, activated macrophage specific chemokine 1, Macrophage inflammatory protein 1 alpha, Macrophage inflammatory protein 1 beta, Macrophage inflammatory protein 1 gamma, Macrophage inflammatory protein 1 delta, Macrophage inflammatory protein 2 alpha, Macrophage inflammatory protein 3 alpha, Macrophage inflammatory protein 3 beta, Regulated upon activation, normal T cell expressed and secreted (and its variants), I-309, EBI1-ligand chemokine, Pulmonary and activation regulated chemokine, Liver and activation-regulated chemokine, Thymus and activation regulated chemokine, Eotaxin (and variants), Human CC chemokine 1, Human CC chemokine 2, Human CC chemokine 3, IL-10-inducible chemokine, liver-expressed chemokine, 6Ckine, Exodus 1, Exodus 2, Exodus 3, thymus-expressed chemokine, Secondary Lymphoid tissue chemokine, Lymphocyte and Monocyte chemoattractant; Monotactin, Activation-induced, chemokine-related molecule, Myeloid progenitor inhibitory factor-1, Myeloid progenitor inhibitory factor-2, Stromal cell-derived factor 1 alpha, Stromal cell-derived factor 1 beta, B-cell-attracting chemokine 1, HuMIG, H174, Interferon-stimulated T-cell alpha chemoattractant, Interleukin-8, IP-10, platelet factor 4, growth-regulated gene-alpha, growth-

regulated gene-beta, growth-regulated gene-gamma, Neutrophil-activating protein 2, ENA-78, granulocyte chemotactic protein 2, LYMPHOTACTIN, and Fractalkine/neurotactin.

4. The method of claim 1, wherein the one or more chemokines are selected from the group consisting of: MDC, SDF-1, BLC, and MCP-1.

5. The method of claim 1 wherein the fragment(s) or derivative(s) are truncation isoforms.

6. The method of claim 1, wherein the one or more chemokines include MDC comprising the amino acid sequence of SEQ ID NO: 2.

7. The method of claim 1, wherein the one or more chemokine fragment includes an MDC fragmentselected from the group consisting of amino acid numbers 2-69, 3-69, 5-69, 7-69 and 9-69 of SEQ ID NO: 2.

8. The method of claim 1, wherein the one or more chemokine fragment includes an MDC fragment selected from the group consisting of amino acid numbers 2-69, 3-69, 5-69, 7-69 and 9-69 of SEQ ID NO: 2., which derivative has activity to enhance the efficacy of the vaccine.

9. The method of claim 1, wherein the one or more chemokine derivatives has one or more insertions or substitutions with one or more non-classical amino acids relative to a corresponding wildtype chemokine, which derivative has activity to enhance the efficacy of the vaccine.

10. The method of claim 1, including a chemokine derivative having one or more conservative substitutions in sequence relative a wildtype MDC, which derivative has activity to enhance the efficacy of the vaccine.

11. The method of claim 1, wherein the one or more chemokines include a human chemokine.

12. The method of claim 1, wherein the purified chemokine(s) or purified fragment(s) or derivative(s) thereof is/are administered concurrently with the purified antigen(s).

13. The method of claim 1 wherein the purified chemokine(s) or purified fragment(s) or derivative(s) thereof, are administered within a time period before or after administration of the purified antigen, which time period permits the purified MDC or purified fragment or derivative thereof MDC to enhance the efficacy of the vaccine.

14. The method of claim 1, wherein the antigen is an HIV antigen.

15. The method of claim 14, wherein the HIV antigen is HIV-associated gp120 protein.

16. The method of claim 1, wherein the subject is a human.

17. The method of claim 1, wherein the subject is infected or at risk of being infected with HIV virus.

18. The method of claim 1, wherein the vaccine elicits a humoral response against the antigen in the subject.

19. The method of claim 1, wherein the vaccine elicits a cell-mediated response against the antigen in the subject.

20. The method of claim 1, wherein the vaccine elicits both a humoral and a cell-mediated response against the antigen in the subject.

21. The method of claim 1, wherein the vaccine further comprises pharmaceutically acceptable excipient, auxiliary substance, adjuvant, wetting or emulsifying agent, or pH buffering agent.

22. A method to enhance the efficacy of a vaccine in a subject comprising administering to the subject a first amount of a first set of one or more purified nucleotide sequences encoding one or more antigens against which an immune response is desired in the subject and a second second set of one or more purified nucleic acids; each comprising a nucleotide sequence encoding one or more chemokines, or fragments or derivatives thereof, wherein the antigen(s) and the chemokine(s) are expressed in a coordinated manner upon introduction into a suitable cell, said first amount is immunogenic and said second amount is effective in enhancing the efficacy of the vaccine.

23. The method of claim 22, wherein the one or more chemokines are selected from a chemokine class selected from the group consisting of: CC, CXC, C-C and CX3C.

24. The method of claim 22, wherein the one or more chemokines are selected from the group consisting of: Macrophage-derived chemokine, Monocyte chemotactic protein 1, Monocyte chemotactic protein 2, Monocyte chemotactic protein 3, Monocyte chemotactic protein 4, activated macrophage specific chemokine 1, Macrophage inflammatory protein 1 alpha, Macrophage inflammatory protein 1 beta, Macrophage inflammatory protein 1 gamma, Macrophage inflammatory protein 1 delta, Macrophage inflammatory protein 2 alpha, Macrophage inflammatory protein 3 alpha, Macrophage inflammatory protein 3 beta, Regulated upon activation, normal T cell expressed and secreted (and its variants), I-309, EBI1-ligand chemokine, Pulmonary and activation regulated chemokine, Liver and activation-regulated chemokine, Thymus and activation regulated chemokine, Eotaxin (and variants), Human CC chemokine 1, Human CC chemokine 2, Human CC chemokine 3, IL-10-inducible chemokine, liver-expressed chemokine, 6Ckine, Exodus 1, Exodus 2, Exodus 3, thymus-expressed chemokine, Secondary Lymphoid tissue chemokine, Lymphocyte and Monocyte chemoattractant; Monotactin, Activation-induced, chemokine-related molecule, Myeloid progenitor inhibitory factor-1, Myeloid progenitor inhibitory factor-2, Stromal cell-derived factor 1 alpha, Stromal cell-derived factor 1 beta, B-cell-attracting chemokine

1, HuMIG, H174, Interferon-stimulated T-cell alpha chemoattractant, Interleukin-8, IP-10, platelet factor 4, growth-regulated gene-alpha, growth-regulated gene-beta, growth-regulated gene-gamma, Neutrophil-activating protein 2, ENA-78, granulocyte chemotactic protein 2, LYMPHOTACTIN, and Fractalkine/neurotactin.

25. The method of claim 22, wherein the one or more chemokines are selected from the group consisting of: MDC, SDF-1, BLC, and MCP-1.

26. The method of claim 22 wherein the fragment(s) or derivative(s) are truncation isoforms.

27. The method of claim 22, wherein the nucleotide sequence encoding one or more chemokines comprises the nucleotide sequence of SEQ ID NO:1.

28. The method of claim 22, wherein one or more of the chemokine derivative(s) have deletional, insertional or substitutional mutations and combination thereof, which derivative has activity to enhance the efficacy of the vaccine.

29. The method of claim 22, wherein the vaccine elicits a humoral response against the antigen in the subject.

30. The method of claim 22, wherein the vaccine elicits a cell-mediated response against the antigen in the subject.

31. The method of claim 22, wherein the vaccine elicits both a humoral and a cell-mediated response against the antigen in the subject.

32. The method of claim 22, wherein the vaccine further comprises pharmaceutically acceptable excipient, auxiliary substance, adjuvant, wetting or emulsifying agent, or pH buffering agent.

33. A composition comprising: an immunogenic amount of one or more purified antigens and an amount of one or more purified chemokines, or purified

fragments or derivatives thereof, effective to enhance the immune response to said antigen(s); and a pharmaceutically acceptable carrier.

34. The composition of claim 33, wherein the one or more chemokines are selected from the group consisting of: MDC, SDF-1, BLC, and MCP-1.

35. The composition of claim 33, wherein the one or more chemokines are selected from a chemokine class selected from the group consisting of: CC, CXC, C-C and CX3C.

36. The composition of claim 33, wherein the one or more chemokines are selected from the group consisting of: Macrophage-derived chemokine, Monocyte chemotactic protein 1, Monocyte chemotactic protein 2, Monocyte chemotactic protein 3, Monocyte chemotactic protein 4, activated macrophage specific chemokine 1, Macrophage inflammatory protein 1 alpha, Macrophage inflammatory protein 1 beta, Macrophage inflammatory protein 1 gamma, Macrophage inflammatory protein 1 delta, Macrophage inflammatory protein 2 alpha, Macrophage inflammatory protein 3 alpha, Macrophage inflammatory protein 3 beta, Regulated upon activation, normal T cell expressed and secreted (and its variants), I-309, EBI1-ligand chemokine, Pulmonary and activation regulated chemokine, Liver and activation-regulated chemokine, Thymus and activation regulated chemokine, Eotaxin (and variants), Human CC chemokine 1, Human CC chemokine 2, Human CC chemokine 3, IL-10-inducible chemokine, liver-expressed chemokine, 6Ckine, Exodus 1, Exodus 2, Exodus 3, thymus-expressed chemokine, Secondary Lymphoid tissue chemokine, Lymphocyte and Monocyte chemoattractant; Monotactin, Activation-induced, chemokine-related molecule, Myeloid progenitor inhibitory factor-1, Myeloid progenitor inhibitory factor-2, Stromal cell-derived factor 1 alpha, Stromal cell-derived factor 1 beta, B-cell-attracting chemokine 1, HuMIG, H174, Interferon-stimulated T-cell alpha chemoattractant, Interleukin-8, IP-10, platelet factor 4, growth-regulated gene-alpha, growth-regulated gene-beta, growth-regulated gene-gamma, Neutrophil-activating

protein 2, ENA-78, granulocyte chemotactic protein 2, LYMPHOTACTIN, and Fractalkine/neurotactin.

37. The composition of claim 33, wherein the fragment(s) or derivative(s) are truncation isoforms.

38. The composition of claim 33, wherein the one or more chemokine fragment includes an MDC fragmentselected from the group consisting of amino acid numbers 2-69, 3-69, 5-69, 7-69 and 9-69 of SEQ ID NO: 2.

39. The composition of claim 33, wherein the one or more chemokine fragment includes an MDC fragmentselected from the group consisting of amino acid numbers 2-69, 3-69, 5-69, 7-69 and 9-69 of SEQ ID NO: 2, which derivative has activity to enhance the efficacy of the vaccine.

40. The composition of claim 33, wherein the one or more chemokine derivatives has one or more insertions of or substitutions with one or more non-classical amino acids relative to a corresponding wildtype chemokine, which derivative has activity to enhance the efficacy of the vaccine.

41. The composition of claim 33, wherein the one or more chemokine derivatives has one or more conservative substitutions in sequence relative a corresponding wildtype chemokine, which derivative has activity to enhance the efficacy of the vaccine.

42. The composition of claim 33, wherein the chemokine is a human chemokine.

43. The composition of claim 33, wherein the antigen is an HIV antigen.

44. The composition of claim 43, wherein the antigen is HIV associated gp120 protein.

45. A composition comprising an amount of a first set of purified nucleic acids comprising one or more nucleotide sequences encoding one or more antigens

and a second set of purified nucleic acids comprising one or more nucleotide sequences encoding one or more chemokines, or fragments or derivatives thereof, wherein the antigen(s) and the chemokine(s), or fragment(s) or derivative(s) thereof, are expressed from said first set of nucleic acid(s) and second set of nucleic acid(s) in a coordinated manner such that upon introduction into a suitable cell, the amount of said first set of nucleic acid(s) is sufficient to express an immunogenic amount of the antigen and the amount of the said second set of nucleic acid(s) is effective in enhancing the efficacy of the vaccine; and a pharmaceutically acceptable carrier.

46. The composition of claim 45, wherein the chemokine is MDC and the nucleic acid encoding the MDC comprises the nucleotide sequence of SEQ ID NO: 1.

47. The composition of claim 45, wherein the chemokine derivative(s) have deletional, insertional or substitutional mutations and/or combinations thereof, and the derivative(s) have activity to enhance the efficacy of the vaccine.

48. The composition of claim 45, further comprising pharmaceutically acceptable excipient, auxiliary substance, adjuvant, wetting or emulsifying agent, or pH buffering agent.

49. A composition comprising a first set of purified nucleotide sequences encoding one or more antigens and a second set of purified nucleotide sequences encoding one or more chemokines, or fragments or derivatives thereof, wherein the antigen(s) and the chemokine(s) are expressed in a coordinated manner such that upon introduction into a suitable cell, the sets produce an amount of said antigen(s) that is immunogenic and an amount of chemokine(s), or fragment(s) or derivative(s) thereof, that is effective in enhancing the efficacy of the vaccine relative to a corresponding vaccine composition without such chemokine(s), fragment(s) or derivative(s) thereof.

50. The composition of claim 49, wherein the one or more chemokines are selected from the group consisting of: Macrophage-derived chemokine,

Monocyte chemotactic protein 1, Monocyte chemotactic protein 2, Monocyte chemotactic protein 3, Monocyte chemotactic protein 4, activated macrophage specific chemokine 1, Macrophage inflammatory protein 1 alpha, Macrophage inflammatory protein 1 beta, Macrophage inflammatory protein 1 gamma, Macrophage inflammatory protein 1 delta, Macrophage inflammatory protein 2 alpha, Macrophage inflammatory protein 3 alpha, Macrophage inflammatory protein 3 beta, Regulated upon activation, normal T cell expressed and secreted (and its variants), I-309, EBI1-ligand chemokine, Pulmonary and activation regulated chemokine, Liver and activation-regulated chemokine, Thymus and activation regulated chemokine, Eotaxin (and variants), Human CC chemokine 1, Human CC chemokine 2, Human CC chemokine 3, IL-10-inducible chemokine, liver-expressed chemokine, 6Ckine, Exodus 1, Exodus 2, Exodus 3, thymus-expressed chemokine, Secondary Lymphoid tissue chemokine, Lymphocyte and Monocyte chemoattractant; Monotactin, Activation-induced, chemokine-related molecule, Myeloid progenitor inhibitory factor-1, Myeloid progenitor inhibitory factor-2, Stromal cell-derived factor 1 alpha, Stromal cell-derived factor 1 beta, B-cell-attracting chemokine 1, HuMIG, H174, Interferon-stimulated T-cell alpha chemoattractant, Interleukin-8, IP-10, platelet factor 4, growth-regulated gene-alpha, growth-regulated gene-beta, growth-regulated gene-gamma, Neutrophil-activating protein 2, ENA-78, granulocyte chemotactic protein 2, LYMPHOTACTIN, and Fractalkine/neurotactin.

51. The method of claim 49, wherein the one or more chemokines are selected from a chemokine class selected from the group consisting of: CC, CXC, C-C and CX3C.

52. The method of claim 49, wherein the one or more chemokines are selected from the group consisting of: MDC, SDF-1, BLC, and MCP-1.

53. The composition of claim 49, wherein the fragment(s) or derivative(s) are truncation isoforms.

54. The composition of claim 49, wherein the nucleic acid is administered directly to the subject.

55. The composition of claim 49, wherein the nucleic acid is introduced into a suitable host cell and said suitable host cell is introduced into the subject.

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GAGACATACA GGACAGAGC ATG GCT CGC CTA CAG ACT GCA CTC CTG GTT GTC       52
                     Met Ala Arg Leu Gln Thr Ala Leu Leu Val Val
                     -24             -20                 -15

CTC GTC CTC CTT GCT GTG GCG CTT CAA GCA ACT GAG GCA GGC CCC TAC       100
Leu Val Leu Leu Ala Val Ala Leu Gln Ala Thr Glu Ala Gly Pro Tyr
            -10                     -5                  1

GGC GCC AAC ATG GAA GAC AGC GTC TGC TGC CGT GAT TAC GTC CGT TAC       148
Gly Ala Asn Met Glu Asp Ser Val Cys Cys Arg Asp Tyr Val Arg Tyr
    5                   10                  15

CGT CTG CCC CTG CGC GTG GTG AAA CAC TTC TAC TGG ACC TCA GAC TCC       196
Arg Leu Pro Leu Arg Val Val Lys His Phe Tyr Trp Thr Ser Asp Ser
20                  25                  30                  35

TGC CCG AGG CCT GGC GTG GTG TTG CTA ACC TTC AGG GAT AAG GAG ATC       244
Cys Pro Arg Pro Gly Val Val Leu Leu Thr Phe Arg Asp Lys Glu Ile
                40                  45                  50

TGT GCC GAT CCC AGA GTG CCC TGG GTG AAG ATG ATT CTC AAT AAG CTG       292
Cys Ala Asp Pro Arg Val Pro Trp Val Lys Met Ile Leu Asn Lys Leu
            55                  60                  65
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FIG. 1A-1

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AGC CAA TGAAGAGCCT ACTCTGATGA CCGTGGCCTT GGCTCCTCCA GGAAGGCTCA      348
Ser Gln

GGAGCCCTAC CTCCCTGCCA TTATAGCTGC TCCCCGCCAG AAGCCTGTGC CAACTCTCTG   408

CATTCCCTGA TCTCCATCCC TGTGGCTGTC ACCCTTGGTC ACCTCCGTGC TGTCACTGCC   468

ATCTCCCCCC TGACCCCTCT AACCCATCCT CTGCCTCCCT CCCTGCAGTC AGAGGGTCCT   528

GTTCCCATCA GCGATTCCCC TGCTTAAACC CTTCCATGAC TCCCCACTGC CCTAAGCTGA   588

GGTCAGTCTC CCAAGCCTGG CATGTGGCCC TCTGGATCTG GGTTCCATCT CTGTCTCCAG   648

CCTGCCCACT TCCCTTCATG AATGTTGGGT TCTAGCTCCC TGTTCTCCAA ACCCATACTA   708

CACATCCCAC TTCTGGGTCT TTGCCTGGGA TGTTGCTGAC ACTCAGAAAG TCCCACCACC   768

TGCACATGTG TAGCCCCACC AGCCCTCCAA GGCATTGCTC GCCCAAGCAG CTGGTAATTC   828

CATTTCATGT ATTAGATGTC CCCTGGCCCT CTGTCCCCTC TTAATAACCC TAGTCACAGT   888

CTCCGCAGAT TCTTGGGATT TGGGGGTTTT CTCCCCCACC TCTCCACTAG TTGGACCAAG   948
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FIG. 1A-2

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GTTTCTAGCT AAGTTACTCT AGTCTCCAAG CCTCTAGCAT AGAGCACTGC AGACAGGCCC   1008
TGGCTCAGAA TCAGAGCCCA GAAAGTGGCT GCAGACAAAA TCAATAAAAC TAATGTCCCT   1068
CCCCTCTCCC TGCCAAAAGG CAGTTACATA TCAATACAGA GACTCAAGGT CACTAGAAAT   1128
GGGCCAGCTG GGTCAATGTG AAGCCCCAAA TTTGCCCAGA TTCACCTTTC TTCCCCCACT   1188
CCCTTTTTTT TTTTTTTTTT TTTGAGATGG AGTTTCGCTC TTGTCACCCA CGCTGGAGTG   1248
CAATGGTGTG GTCTTGGCTT ATTGAAGCCT CTGCCTCCTG GGTTCAAGTG ATTCTCTTGC   1308
CTCAGCCTCC TGAGTAGCTG GGATTACAGG TTCCTGCTAC CACGCCCAGC TAATTTTTGT   1368
ATTTTTAGTA GAGACGAGGC TTCACCATGT TGGCCAGGCT GGTCTCGAAC TCCTGTCCTC   1428
AGGTAATCCG CCCACCTCAG CCTCCCAAAG TGCTGGGATT ACAGGCGTGA GCCACAGTGC   1488
CTGGCCTCTT CCCTCTCCCC ACTGCCCCCC CCAACTTTTT TTTTTTTTTT ATGGCAGGGT   1548
CTCACTCTGT CGCCCAGGCT GGAGTGCAGT GGCGTGATCT CGGCTCACTA CAACCTCGAC   1608
CTCCTGGGTT CAAGTGATTC TCCCACCCCA GCCTCCCAAG TAGCTGGGAT TACAGGTGTG   1668
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FIG. 1A-3

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TGCCACTACG GCTGGCTAAT TTTTGTATTT TTAGTAGAGA CAGGTTTCAC CATATTGGCC  1728
AGGCTGGTCT TGAACTCCTG ACCTCAAGTG ATCCACCTTC CTTGTGCTCC CAAAGTGCTG  1788
AGATTACAGG CGTGAGCTAT CACACCCAGC CTCCCCCTTT TTTTCCTAAT AGGAGACTCC  1848
TGTACCTTTC TTCGTTTTAC CTATGTGTCG TGTCTGCTTA CATTTCCTTC TCCCCTCAGG  1908
CTTTTTTTGG GTGGTCCTCC AACCTCCAAT ACCCAGGCCT GGCCTCTTCA GAGTACCCCC  1968
CATTCCACTT TCCCTGCCTC CTTCCTTAAA TAGCTGACAA TCAAATTCAT GCTATGGTGT  2028
GAAAGACTAC CTTTGACTTG GTATTATAAG CTGGAGTTAT ATATGTATTT GAAAACAGAG  2088
TAAATACTTA AGAGGCCAAA TAGATGAATG GAAGAATTTT AGGAACTGTG AGAGGGGGAC  2148
AAGGTGAAGC TTTCCTGGCC CTGGGAGGAA GCTGGCTGTG GTAGCGTAGC GCTCTCTCTC  2208
TCTGTCTGTG GCAGGAGCCA AAGAGTAGGG TGTAATTGAG TGAAGGAATC CTGGGTAGAG  2268
ACCATTCTCA GGTGGTTGGG CCAGGCTAAA GACTGGGAGT TGGGTCTATC TATGCCTTTC  2328
TGGCTGATTT TTGTAGAGAC GGGGTTTTGC CATGTTACCC AGGCTGGTCT CAAACTCCTG  2388
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FIG. 1A-4

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GGCTCAAGCG ATCCTCCTGG CTCAGCCTCC CAAAGTGCTG GGATTACAGG CGTGAATCAC    2448
TGCGCCTGGC TTCCTCTTCC TCTTGAGAAA TATTCTTTTC ATACAGCAAG TATGGGACAG    2508
CAGTGTCCCA GGTAAAGGAC ATAAATGTTA CAAGTGTCTG GTCCTTTCTG AGGGAGGCTG    2568
GTGCCGCTCT GCAGGGTATT TGAACCTGTG GAATTGGAGG AGGCCATTTC ACTCCCTGAA    2628
CCCAGCCTGA CAAATCACAG TGAGAATGTT CACCTTATAG GCTTGCTGTG GGGCTCAGGT    2688
TGAAAGTGTG GGGAGTGACA CTGCCTAGGC ATCCAGCTCA GTGTCATCCA GGGCCTGTGT    2748
CCCTCCCGAA CCCAGGGTCA ACCTGCCTGC CACAGGCACT AGAAGGACGA ATCTGCCTAC    2808
TGCCCATGAA CGGGGCCCTC AAGCGTCCTG GGATCTCCTT CTCCCTCCTG TCCTGTCCTT    2868
GCCCCTCAGG ACTGCTGGAA AATAAATCCT TTAAAATAGT AAAAAAAAAA AAAAA         2923
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FIG. 1A-5

FIG. 1A-1
FIG. 1A-2
FIG. 1A-3
FIG. 1A-4
FIG. 1A-5

FIG. 1A

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Met Ala Arg Leu Gln Thr Ala Leu Leu Val Val Leu Val Leu Leu Ala
-24             -20                 -15                 -10
Val Ala Leu Gln Ala Thr Glu Ala Gly Pro Tyr Gly Ala Asn Met Glu
             -5                   1               5
Asp Ser Val Cys Cys Arg Asp Tyr Val Arg Tyr Arg Leu Pro Leu Arg
     10                  15                  20
Val Lys His Phe Tyr Tyr Trp Thr Ser Asp Ser Cys Pro Arg Pro Gly
 25                  30                  35                  40
Val Val Leu Leu Thr Phe Arg Asp Lys Glu Ile Cys Ala Asp Pro Arg
                 45                  50                  55
Val Pro Trp Val Lys Met Ile Leu Asn Lys Leu Ser Gln
             60                  65
```

FIG. 1B

SEQUENCE LISTING

(1) GENERAL INFORMATION

(i) APPLICANT: Gallo, Robert C.
DeVico, Anthony L.
Garzino, Alfedo

(ii) TITLE OF THE INVENTION: METHOD AND COMPOSITION TO ENHANCE THE EFFICACY OF A VACCINE USING MACROPHAGE DERIVED CHEMOKINE

(iii) NUMBER OF SEQUENCES: 2
(iv) CORRESPONDENCE ADDRESS:
(A) ADDRESSEE: Pennie & Edmonds LLP
(B) STREET: 1155 Avenue of the Americas
(C) CITY: New York
(D) STATE: New York
(E) COUNTRY: USA
(F) ZIP: 10036/2711
(v) COMPUTER READABLE FORM:
(A) MEDIUM TYPE: Diskette
(B) COMPUTER: IBM Compatible
(C) OPERATING SYSTEM: DOS
(D) SOFTWARE: FastSEQ Version 2.0
(vi) CURRENT APPLICATION DATA:
(A) APPLICATION NUMBER: To be assigned
(B) FILING DATE: Herewith
(C) CLASSIFICATION:
(viii) ATTORNEY/AGENT INFORMATION:
(A) NAME: Misrock, S. Leslie
(B) REGISTRATION NUMBER: 18,872
(C) REFERENCE/DOCKET NUMBER: 8769-029
(ix) TELECOMMUNICATION INFORMATION:
(A) TELEPHONE: 212-790-9090
(B) TELEFAX: 212-869-8864
(C) TELEX: 66141 PENNIE
(2) INFORMATION FOR SEQ ID NO:1:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 2923 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear
(ii) MOLECULE TYPE: DNA

(ix) FEATURE:
(A) NAME/KEY: mat_peptide
(B) LOCATION: 92..298

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:1:

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GAGACATACA GGACAGAGC ATG GCT CGC CTA CAG ACT GCA CTC CTG GTT GTC        52
                     Met Ala Arg Leu Gln Thr Ala Leu Val
                     -24             -20                 -15
CTC GTC CTC CTT GCT GTG GCG CTT CAA GCA ACT GAG GCA GGC CCC TAC        100
Leu Val Leu Ala Val Ala Leu Gln Ala Thr Glu Ala Gly Pro Tyr
        -10                 -5                  1
GGC GCC AAC ATG GAA GAC AGC GTC TGC CGT GAT TAC GTC CGT TAC        148
Gly Ala Asn Met Glu Asp Ser Val Cys Arg Asp Tyr Val Arg Tyr
    5                   10                  15
CGT CTG CCC CTG CGC GTG AAA CAC TTC TAC TGG ACC TCA GAC TCC        196
Arg Leu Pro Leu Arg Val Lys His Phe Tyr Trp Thr Ser Asp Ser
 20                 25                  30                  35

TGC CCG AGG CCT GGC GTG TTG CTA ACC TTC AGG GAT AAG GAG ATC        244
Cys Pro Arg Pro Gly Val Leu Thr Phe Arg Asp Lys Glu Ile
            40                  45                  50
TGT GCC GAT CCC AGA GTG CCC TGG GTG AAG ATG ATT CTC AAT AAG CTG        292
Cys Ala Asp Pro Arg Val Pro Trp Val Lys Met Ile Leu Asn Lys Leu
        55                  60                  65
AGC CAA TGAAGAGCCT ACTCTGATGA CCGTGGCCTT GGCTCCTCCA GGAAGGCTCA         348
Ser Gln
```

GGAGCCCTAC CTCCCTGCCA TTATAGCTGC TCCCCGCCAG AAGCCTGTGC CAACTCTCTG 408
CATTCCCTGA TCTCCATCCC TGTGGCTGTC ACCCTTGGTC ACCTCCGTGC TGTCACTGCC 468
ATCTCCCCCC TGACCCCTCT AACCCATCCT CTGCCTCCCT CCCTGCAGTC AGAGGGTCCT 528
GTTCCCATCA GCGATTCCCC TGCTTAAACC CTTCCATGAC TCCCCACTGC CCTAAGCTGA 588
GGTCAGTCTC CCAAGCCTGG CATGTGGCCC TCTGGATCTG GGTTCCATCT CTGTCTCCAG 648
CCTGCCCACT TCCCTTCATG AATGTTGGGT TCTAGCTCCC TGTTCTCCAA ACCCATACTA 708
CACATCCCAC TTCTGGGTCT TTGCCTGGGA TGTTGCTGAC ACTCAGAAAG TCCCACCACC 768
TGCACATGTG TAGCCCCACC AGCCCTCCAA GGCATTGCTC GCCCAAGCAG CTGGTAATTC 828
CATTTCATGT ATTAGATGTC CCCTGGCCCT CTGTCCCCTC TTAATAACCC TAGTCACAGT 888
CTCCGCAGAT TCTTGGGATT TGGGGGTTTT CTCCCCCACC TCTCCACTAG TTGGACCAAG 948
GTTTCTAGCT AAGTTACTCT AGTCTCCAAG CCTCTAGCAT AGAGCACTGC AGACAGGCCC 1008
TGGCTCAGAA TCAGAGCCCA GAAAGTGGCT GCAGACAAAA TCAATAAAAC TAATGTCCCT 1068
CCCCTCTCCC TGCCAAAAGG CAGTTACATA TCAATACAGA GACTCAAGGT CACTAGAAAT 1128
GGGCCAGCTG GGTCAATGTG AAGCCCCAAA TTTGCCCAGA TTCACCTTTC TTCCCCCACT 1188
CCCTTTTTTT TTTTTTTTTT TTTGAGATGG AGTTTCGCTC TTGTCACCCA CGCTGGAGTG 1248
CAATGGTGTG GTCTTGGCTT ATTGAAGCCT CTGCCTCCTG GGTTCAAGTG ATTCTCTTGC 1308
CTCAGCCTCC TGAGTAGCTG GGATTACAGG TTCCTGCTAC CACGCCCAGC TAATTTTTGT 1368
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(2) INFORMATION FOR SEQ ID NO:2:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 93 amino acids
(B) TYPE: amino acid
(D) TOPOLOGY: linear
(ii) MOLECULE TYPE: protein
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

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Asp Ser Val Cys Arg Asp Tyr Val Arg Tyr Arg Leu Pro Leu Arg
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C-CHEMOKINES
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CX3C-chemokines
Fractalkine/neurotactin U91835 U84487

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DEFINITION    Human  macrophage-derived  chemokine  precursor  (MDC)  mRNA,
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ACCESSION   U83171
NID         g1931580
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
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            Vertebrata;    Mammalia;    Eutheria;    Primates;    Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (bases 1 to 2923)
  AUTHORS   Godiska,R., Chantry,D., Raport,C.J., Sozzani,S., Allavena,P.,
            Leviten,D., Mantovani,A. and Gray,P.W.
  TITLE            Human   macrophage-derived   chemokine   (MDC),   a   novel
chemoattractant
            for monocytes, monocyte-derived dendritic cells, and natural
killer
            cells
  JOURNAL   J. Exp. Med. 185 (9), 1595-1604 (1997)
  MEDLINE   97296313
REFERENCE   2  (bases 1 to 2923)
  AUTHORS   Godiska,R. and Gray,P.W.
  TITLE     Direct Submission
  JOURNAL   Submitted (23-DEC-1996) ICOS Corporation, 22021 20th Avenue SE,
            Bothell, WA 98021, USA
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LOCUS       HSU83239      932 bp    mRNA            PRI       02-MAY-1997
DEFINITION  Human CC chemokine STCP-1 mRNA, complete cds.
ACCESSION   U83239
NID         g2062424
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 932)
  AUTHORS       Chang,M.S.,    McNinch,J.,    Elias    III,C.,    Manthey,C.L.,
Grosshans,D.,
            Meng,T., Boone,T. and Andrew,D.P.
  TITLE     Molecular cloning and functional characterization of a novel CC
            chemokine  STCP-1  which  specifically  acts  on  activated  T
lymphocytes
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 932)
  AUTHORS       Chang,M.S.,    McNinch,J.,    Elias    III,C.,    Manthey,C.L.,
Grosshans,D.,
            Meng,T., Boone,T. and Andrew,D.P.
  TITLE     Direct Submission
  JOURNAL    Submitted (26-DEC-1996) Research Computing, Amgen Institute,
620
            University Ave, Suite 706, Toronto, ON M5G 2C1, Canada
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DEFINITION  H.sapiens mRNA for monocyte chemoattractant protein 1 (MCP-1).
ACCESSION   X14768
NID         g34513
KEYWORDS    monocyte chemoattractant protein 1.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 725)
  AUTHORS   Yoshimura,T., Yuhki,N., Moore,S.K., Appella,E., Lerman,M.I. and
            Leonard,E.J.
  TITLE      Human monocyte chemoattractant protein-1 (MCP-1). Full-length
cDNA
            cloning, expression in mitogen-stimulated blood mononuclear
            leukocytes, and sequence similarity to mouse competence gene JE
  JOURNAL   FEBS Lett. 244 (2), 487-493 (1989)
  MEDLINE   89153605
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DEFINITION  H.sapiens MCP-2 gene.
ACCESSION   X99886
NID         g1905800
KEYWORDS    MCP-2 gene; monocyte chemotactic protein 2; SCYA10 gene.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 2991)
  AUTHORS   Van Coillie,E., Fiten,P., Nomiyama,H., Sakaki,Y., Miura,R.,
            Yoshie,O., Van Damme,J. and Opdenakker,G.
  TITLE       The human MCP-2 gene (SCYA8): cloning, sequence analysis,
tissue
            expression, and assignment to the CC chemokine gene contig on
            chromosome 17q11.2
  JOURNAL   Genomics 40 (2), 323-331 (1997)
  MEDLINE   97237052
REFERENCE   2  (bases 1 to 2991)
  AUTHORS   Opdenakker,G.M.M.
  TITLE     Direct Submission
  JOURNAL   Submitted (07-AUG-1996) G.M.M. Opdenakker, Rega Institute for
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BELGIUM
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SOURCE      human.
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            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1368)
  AUTHORS   Van Coillie,E.
  TITLE       Functional comparison of two human monocyte chemotactic
protein-2
            isoforms, role of the amino-terminal pyroglutamic acid and
            processing by CD26/dipeptidyl peptidase IV
  JOURNAL   Biochemistry 37, 12672-12680 (1998)
REFERENCE   2  (bases 1 to 1368)
  AUTHORS   Van Coillie,E.
  TITLE     Direct Submission
  JOURNAL     Submitted (23-FEB-1998) E. Van Coillie, Rega Institute for
Medical
            Research, Minderbroedersstraat 10, 3000 Leuven, BELGIUM
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ACCESSION   X72308 S57464
NID         g313707
KEYWORDS    monocyte chemotactic protein 3.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1085)
  AUTHORS   Opdenakker,G., Froyen,G., Fiten,P., Proost,P. and Van Damme,J.
  TITLE     Human monocyte chemotactic protein-3 (MCP-3): molecular cloning
of
            the cDNA and comparison with other chemokines
  JOURNAL   Biochem. Biophys. Res. Commun. 191 (2), 535-542 (1993)
  MEDLINE   93213290
REFERENCE   2  (bases 1 to 1085)
  AUTHORS   Opdenakker,G.M.
  TITLE     Direct Submission
  JOURNAL      Submitted  (27-MAY-1993)  G.M.  Opdenakker,  Rega  Institute,
University
            of Leuven, Minderbroedersstraat 10, B-3000 Leuven, BELGIUM
REFERENCE   3  (bases 1 to 1085)
  AUTHORS   Opdenakker,G., Fiten,P., Nys,G., Froyen,G., Van Roy,N.,
            Speleman,F., Laureys,G. and Van Damme,J.
  TITLE     The human MCP-3 gene (SCYA7): cloning, sequence analysis, and
            assignment to the C-C chemokine gene cluster on chromosome
            17q11.2-q12
  JOURNAL   Genomics 21 (2), 403-408 (1994)
  MEDLINE   94375065
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LOCUS       HSMCP3A       1085 bp    DNA             PRI       25-JUL-1994
DEFINITION  H.sapiens MCP-3 mRNA for monocyte chemotactic protein-3.
ACCESSION   X72308 S57464
NID         g313707
KEYWORDS    monocyte chemotactic protein 3.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1085)
  AUTHORS   Opdenakker,G., Froyen,G., Fiten,P., Proost,P. and Van Damme,J.
  TITLE     Human monocyte chemotactic protein-3 (MCP-3): molecular cloning
of
            the cDNA and comparison with other chemokines
  JOURNAL   Biochem. Biophys. Res. Commun. 191 (2), 535-542 (1993)
  MEDLINE   93213290
REFERENCE   2  (bases 1 to 1085)
  AUTHORS   Opdenakker,G.M.
  TITLE     Direct Submission
  JOURNAL     Submitted  (27-MAY-1993)  G.M.  Opdenakker,  Rega  Institute,
University
            of Leuven, Minderbroedersstraat 10, B-3000 Leuven, BELGIUM
REFERENCE   3  (bases 1 to 1085)
  AUTHORS   Opdenakker,G., Fiten,P., Nys,G., Froyen,G., Van Roy,N.,
            Speleman,F., Laureys,G. and Van Damme,J.
  TITLE     The human MCP-3 gene (SCYA7): cloning, sequence analysis, and
            assignment to the C-C chemokine gene cluster on chromosome
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  JOURNAL   Genomics 21 (2), 403-408 (1994)
  MEDLINE   94375065
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     1081 aaaaa
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DEFINITION  Human monocyte chemoattractant protein-4 precursor (MCP-4)
mRNA,
            complete cds.
ACCESSION   U46767
NID         g1732122
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 825)
  AUTHORS       Garcia-Zepeda,E.A.,    Combadiere,C.C.,    Rothenberg,M.E.,
Sarafi,M.N.,
            Lavigne,F., Hamid,Q., Murphy,P. and Luster,A.D.
  TITLE     Human monocyte chemoattractant Protein (MCP)-4: A novel CC
            chemokine  with  activities  on  monocytes,  eosinophils,  and
basophils
            induced in allergic and non-allergic inflammation that signals
            through the CC chemokine receptors CCR-2 and 3
  JOURNAL   J. Immunol. 158 (1996) In press
REFERENCE   2  (bases 1 to 825)
  AUTHORS   Garcia-Zepeda,E.A. and Luster,A.D.
  TITLE     Direct Submission
  JOURNAL   Submitted (22-JAN-1996) Eduardo A. Garcia-Zepeda, Infectious
            Disease Unit, Massachusets General Hospital, 149 13th St.,
            Charlestown, MA 02129, USA
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DEFINITION  Homo sapiens mRNA for alternative activated macrophage specific
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ACCESSION   Y13710
NID         g2326515
KEYWORDS    AMAC-1 gene; CC-chemokine 1.
SOURCE      human. ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (bases 1 to 803)
  AUTHORS   Politz,O.
  TITLE     Direct Submission
  JOURNAL   Submitted (10-JUN-1997) Politz O., Dermatology, Free University
            Benjamin  Franklin  Medical  Center,  Hindenburgdamm  30;  12200
Berlin
            GERMANY
REFERENCE   2  (bases 1 to 803)
  AUTHORS   Kodelja,V., Mueller,C., Politz,O., Hakiy,N., Orfanos,C.E. and
            Goerdt,S.
  TITLE       Cloning  of  alternative  activated  macrophage  associated  CC
chemokine
            1 (AMAC-1)
  JOURNAL   Unpublished
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DEFINITION  Human LD78 alpha gene.
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KEYWORDS    LD78; LD78 alpha; cytokine; inducible gene family; secreted
            peptide.
SOURCE      Human blood lymphocyte DNA, clone Lm LD-3.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (bases 1 to 3176)
  AUTHORS   Nakao,M., Nomiyama,H. and Shimada,K.
  TITLE     Structures of human genes coding for cytokine LD78 and their
            expression
  JOURNAL   Mol. Cell. Biol. 10 (7), 3646-3658 (1990)
  MEDLINE   90287155
COMMENT       These data kindly submitted in computer readable form by:
Hisayuki
            Nomiyama
            Department of Biochemistry
            Kumamoto University Medical School
            2-2-1 Honjo, Kumamoto 860
            Japan
            Phone:  096-344-2111
            Fax:    096-372-6140.
FEATURES             Location/Qualifiers
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                     /db_xref="taxon:9606"
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DEFINITION   Homo sapiens macrophage inflammatory protein 1 alpha (MIP1a)
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            partial cds.
ACCESSION   AF043339
NID         g2905627
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 225)
  AUTHORS   Jang,J.S. and Kim,B.E.
  TITLE     Direct Submission
  JOURNAL    Submitted (15-JAN-1998) Protein Engineering, General Institute
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of
            Technology, Hyundai Pharm. Ind. Co., Ltd., 213 Sosa Bon 1-dong,
            Sosa-gu, Bucheon 422-231, Korea
COMMENT     forward primer (5'-tgcgcatcacttgctgctgaca-3')
            reverse primer (5'-cttctggacccctcaggcact-3').
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LOCUS       HUMLD78B      3112 bp    DNA              PRI       17-JAN-1992
DEFINITION  Human LD78 beta gene.
ACCESSION   D90145
NID         g219907
KEYWORDS      LD78;  LD78  beta;  cytokine;  inducible  gene  family;  secreted
peptide.
SOURCE      Human placenta DNA, clone Lm LD-1.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;    Eutheria;    Primates;    Catarrhini;
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            Homo.
REFERENCE   1  (bases 1 to 3112)
  AUTHORS   Nakao,M., Nomiyama,H. and Shimada,K.
  TITLE     Structures of human genes coding for cytokine LD78 and their
            expression
  JOURNAL   Mol. Cell. Biol. 10 (7), 3646-3658 (1990)
  MEDLINE   90287155
COMMENT       These  data  kindly  submitted  in  computer  readable  form  by:
Hisayuki
            Nomiyama
            Department of Biochemistry
            Kumamoto University Medical School
            2-2-1 Honjo, Kumamoto 860
            Japan
            Phone:   096-344-2111
            Fax:     096-372-6140.
FEATURES             Location/Qualifiers
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LOCUS       HUMACT2A      696 bp    mRNA            PRI       30-OCT-1994
DEFINITION  Human activation (Act-2) mRNA, complete cds.
ACCESSION   J04130
NID         g178017
KEYWORDS    act2 gene; immune activation gene.
SOURCE      Human (Hut-102B2 library) activated T cells, cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 696)
  AUTHORS      Lipes,M.A.,   Napolitano,M.,   Jeang,K.T.,   Chang,N.T.   and
Leonard,W.J.
  TITLE     Identification, cloning, and characterization of an immune
            activation gene
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 85 (24), 9704-9708 (1988)
  MEDLINE   89071764
COMMENT     Draft entry and computer-readable sequence [1] kindly submitted
by
            W.Leonard, 09-JAN-1989.
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      661 ccataataaa actttaaaat aaaatgcaga cagtta
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LOCUS       AF031587      481 bp    mRNA            PRI       02-JAN-1998
DEFINITION  Homo sapiens MIP-1 delta mRNA, complete cds.
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ACCESSION   AF031587
NID         g2739163
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 481)
  AUTHORS   Wang,W.
  TITLE      Molecular cloning and characterization of a new CC chemokine
MIP-1
            delta
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 481)
  AUTHORS   Wang,W.
  TITLE     Direct Submission
  JOURNAL   Submitted (27-OCT-1997) Immunobiology, DNAX Research Institute,
901
            California Ave, Palo Alto, CA 94304, USA
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LOCUS       AF043340      234 bp    mRNA             PRI       23-FEB-1998
DEFINITION   Homo sapiens macrophage inflammatory protein 2 alpha (MIP2a)
mRNA,
            partial cds.
ACCESSION   AF043340
NID         g2905629
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 234)
  AUTHORS   Jang,J.S. and Kim,B.E.
  TITLE     Direct Submission
  JOURNAL    Submitted (15-JAN-1998) Protein Engineering, General Institute
of
            Technology, Hyundai Pharm. Ind. Co., Ltd., 213 Sosa Bon 1-dong,
            Sosa-gu, Bucheon 422-231, Korea
COMMENT     forward primer (5'-tgcgcacccctggccactgaactg-3')
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FEATURES             Location/Qualifiers
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                     /db_xref="taxon:9606"
                     /cell_type="PHA-treated peripheral blood leukocyte"
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     gene            <1..234
                     /gene="MIP2a"
     primer_bind     <1..21
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cycles;
                     DeltaCycler II from Ericomp"
     CDS             <1..222
                     /gene="MIP2a"
                     /function="CXC chemokine"
                     /function="proinflammatory cytokine involved in
                     inflammation"
                     /note="8-10 kDa"
                     /codon_start=1
                     /product="macrophage inflammatory protein 2 alpha"
                     /db_xref="PID:g2905630"

/translation="APLATELRCQCLQTLQGIHLKNIQSVKVKSPGPHCAQTEVIATL
                     KNGQKACLNPASPMVKKIIEKMLKNGKSN"
     primer_bind     complement(214..234)
                     /gene="MIP2a"
BASE COUNT       74 a     70 c     54 g     36 t
ORIGIN
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       61 aagaacatcc aaagtgtgaa ggtgaagtcc cccggacccc actgcgccca aaccgaagtc
      121 atagccacac tcaagaatgg gcagaaagct tgtctcaacc ccgcatcgcc catggttaag
      181 aaaatcatcg aaaagatgct gaaaaatggc aaatccaact gaccagaagg aagg
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LOCUS       HSU77035     764 bp    mRNA            PRI       23-JAN-1997
DEFINITION  Human macrophage inflammatory protein 3 alpha (MIP-3a) mRNA,
            complete cds.
ACCESSION   U77035
NID         g1790924
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 764)
  AUTHORS   Rossi,D.L., Vicari,A.P., Franz-Bacon,K., McClanahan,T.K. and
            Zlotnik,A.
  TITLE     Identification through bioinformatics of two new macrophage
            proinflammatory human chemokines: MIP-3alpha and MIP-3beta
  JOURNAL   J. Immunol. 158 (3), 1033-1036 (1997)
  MEDLINE   97166046
REFERENCE   2  (bases 1 to 764)
  AUTHORS   Rossi,D.L. and Zlotnik,A.
  TITLE     Direct Submission
  JOURNAL    Submitted (31-OCT-1996) Immunology, DNAX Research Institute,
901
            California Ave., Palo Alto, CA 94304, USA
FEATURES             Location/Qualifiers
     source          1..764
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
                     /cell_type="elutriated monocytes activated with
                     LPS/IFN-GAMMA"
     gene            1..291
                     /gene="MIP-3a"
     CDS             1..291
                     /gene="MIP-3a"
                     /note="chemokine"
                     /codon_start=1
                     /product="macrophage inflammatory protein 3 alpha"
                     /db_xref="PID:g1790925"

/translation="MCCTKSLLLAALMSVLLLHLCGESEAASNFDCCLGYTDRILHPK
                     FIVGFTRQLANEGCDINAIIFHTKKKLSVCANPKQTWVKYIVRLLSKKVKNM"
BASE COUNT      235 a    121 c    146 g    260 t      2 others
ORIGIN
        1 atgtgctgta ccaagagttt gctcctggct gctttgatgt cagtgctgct actccacctc
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      61 tgcggcgaat cagaagcagc aagcaacttt gactgctgtc ttggatacac agaccgtatt
     121 cttcatccta aatttattgt gggcttcaca cggcagctgg ccaatgaagg ctgtgacatc
     181 aatgctatca tctttcacac aaagaaaaag ttgtctgtgt gcgcaaatcc aaaacagact
     241 tgggtgaaat atattgtgcg tctcctcagt aaaaaagtca agaacatgta aaaactgtgg
     301 cttttctgga atggaattgg acatagccca agaacagaaa gaaccttgct ggggttggag
     361 gtttcacttg cacatcatgg agggtttagt gcttatctaa tttgtgcctc actggacttg
     421 tccaattaat gaagttgatt catattgcat catagtttgc tttgtttaag catcacatta
     481 aagttaaact gtattttatg ttatttatag ctgtaggttt tctgtgttta gctatttaat
     541 actaattttc cataagctat tttggtttag tgcaaagtat aaaattatat ttgggggga
     601 ataagattat atggactttt ttgcaagcaa caagctattt tttaaaamma actatttaac
     661 attcttttgt ttatattgtt ttgtctccta aattgttgta attgcattat aaaataagaa
     721 aaatattaat aagacaaata ttgaaaataa agaaacaaaa agtt
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LOCUS       HSU77180      545 bp    mRNA            PRI       23-JAN-1997
DEFINITION  Human macrophage inflammatory protein 3 beta (MIP-3beta) mRNA,
            complete cds.
ACCESSION   U77180
NID         g1791002
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 545)
  AUTHORS   Rossi,D.L., Vicari,A.P., Franz-Bacon,K., McClanahan,T.K. and
            Zlotnik,A.
  TITLE     Identification through bioinformatics of two new macrophage
            proinflammatory human chemokines: MIP-3alpha and MIP-3beta
  JOURNAL   J. Immunol. 158 (3)., 1033-1036 (1997)
  MEDLINE   97166046
REFERENCE   2  (bases 1 to 545)
  AUTHORS   Vicari,A. and Zlotnik,A.
  TITLE     Direct Submission
  JOURNAL    Submitted (01-NOV-1996) Immunology, DNAX Research Institute,
901
            California Ave, Palo Alto, CA 94304, USA
FEATURES             Location/Qualifiers
     source          1..545
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
                     /cell_type="macrophages activated with LPS or IFNg"
                     /chromosome="9"
     gene            1..297
                     /gene="MIP-3beta"
     CDS             1..297
                     /gene="MIP-3beta"
                     /function="chemokine"
                     /codon_start=1
                     /product="macrophage inflammatory protein 3 beta"
                     /db_xref="PID:g1791003"

/translation="MALLLALSLLVLWTSPAPTLSGTNDAEDCCLSVTQKPIPGYIVR

NFHYLLIKDGCRVPAVVFTTLRGRQLCAPPDQPWVERIIQRLQRTSAKMKRRSS"
BASE COUNT      125 a    160 c    153 g    107 t
ORIGIN
       1 atggccctgc tactggccct cagcctgctg gttctctgga cttccccagc cccaactctg
      61 agtggcacca atgatgctga agactgctgc ctgtctgtga cccagaaacc catccctggg
     121 tacatcgtga ggaacttcca ctaccttctc atcaaggatg gctgcagggt gcctgctgta
     181 gtgttcacca cactgagggg ccgccagctc tgtgcacccc cagaccagcc ctgggtagaa
     241 cgcatcatcc agagactgca gaggacctca gccaagatga agcgccgcag cagttaacct
     301 atgaccgtgc agagggagcc cggagtccga gtcaagcatt gtgaattatt acctaacctg
     361 gggaaccgag gaccagaagg aaggaccagg cttccagctc ctctgcacca gacctgacca
     421 gccaggacag ggcctggggt gtgtgtgagt gtgagtgtga gcgagagggt gagtgtggtc
     481 tagagtaaag ctgctccacc cccagattgc aatgctacca ataaagccgc ctggtgttta
     541 caact
//
LOCUS       HUMTCSM      1160 bp    mRNA            PRI       15-JUN-1989
DEFINITION  Human T cell-specific protein (RANTES) mRNA, complete cds.
ACCESSION   M21121
NID         g339420
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KEYWORDS      Alu repeat; T-cell-specific protein.
SOURCE          Human peripheral blood (T lymphocyte) cell line AH2, cDNA to
mRNA,
              clone 228.
  ORGANISM    Homo sapiens
              Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
              Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE     1  (bases 1 to 1160)
  AUTHORS           Schall,T.J.,   Jongstra,J.,   Dyer,B.J.,   Jorgensen,J.,
Clayberger,C.,
              Davis,M.M. and Krensky,A.M.
  TITLE         A human T cell-specific molecule is a member of a new gene
family
  JOURNAL     J. Immunol. 141, 1018-1025 (1988)
  MEDLINE     88285659
COMMENT         Draft entry and computer-readable sequence for [1] kindly
provided
              by A.M.Krensky, 24-OCT-1988.
FEATURES             Location/Qualifiers
     source          1..1160
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                     /db_xref="taxon:9606"
     CDS             27..302
                     /note="T cell-specific protein precursor"
                     /codon_start=1
                     /db_xref="PID:g339421"

/translation="MKVSAARLAVILIATALCAPASASPYSSDTTPCCFAYIARPLPR
                     AHIKEYFYTSGKCSNPAVVFVTRKNRQVCANPEKKWVREYINSLEMS"
     sig_peptide     27..95
                     /note="T cell-specific protein signal peptide"
     mat_peptide     96..299
                     /note="T cell-specific protein"
     repeat_region   450..950
                     /note="Alu-related repeats"
BASE COUNT      298 a    332 c    295 g    235 t
ORIGIN        276 bp upstream of RsaI site.
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       61 tcattgctac tgccctctgc gctcctgcat ctgcctcccc atattcctcg gacaccacac
      121 cctgctgctt tgcctacatt gcccgcccac tgccccgtgc ccacatcaag gagtatttct
      181 acaccagtgg caagtgctcc aacccagcag tcgtctttgt cacccgaaag aaccgccaag
      241 tgtgtgccaa cccagagaag aaatgggttc gggagtacat caactctttg gagatgagct
      301 aggatggaga gtccttgaac ctgaacttac acaaatttgc ctgtttctgc ttgctcttgt
      361 cctagcttgg gaggcttccc ctcactatcc taccccaccc gctccttgaa gggcccagat
      421 tctgaccacg acgagcagca gttacaaaaa ccttccccag gctggacgtg gtggctcagc
      481 cttgtaatcc cagcactttg ggaggccaag gtgggtggat cacttgaggt caggagttcg
      541 agacagcctg gccaacatga tgaaaccccа tgtgtactaa aaatacaaaa aattagccgg
      601 gcgtggtagc gggcgcctgt agtcccagct actcgggagg ctgaggcagg agaatggcgt
      661 gaacccggga gcggagcttg cagtgagccg agatcgcgcc actgcactcc agcctgggcg
      721 acagagcgag actccgtctc aaaaaaaaaa aaaaaaaaaa aaaaaataca aaaattagcc
      781 gcgtggtggc ccacgcctgt aatcccagct actcgggagg ctaaggcagg aaaattgttt
      841 gaacccagga ggtggaggct gcagtgagct gagattgtgc cacttcactc cagcctgggt
      901 gacaaagtga gactccgtca caacaacaac aacaaaaagc ttccccaact aaagcctaga
      961 agagcttctg aggcgctgct ttgtcaaaag gaagtctcta ggttctgagc tctggctttg
     1021 ccttggcttt gcaagggctc tgtgacaagg aaggaagtca gcatgcctct agaggcaagg
     1081 aagggaggaa cactgcactc ttaagcttcc gccgtctcaa cccctcacag gagcttactg
     1141 gcaaacatga aaaatcgggg
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LOCUS       HUMTLI309      520 bp    mRNA            PRI       14-JAN-1995
DEFINITION  Human secreted protein (I-309) mRNA, complete cds.
ACCESSION   M57502
NID         g339728
KEYWORDS    secreted protein.
SOURCE      Human T-cell, cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 520)
  AUTHORS   Miller,M.D., Hata,S., De Waal Malefyt,R. and Krangel,M.S.
  TITLE     A novel polypeptide secreted by activated human T lymphocytes
  JOURNAL   J. Immunol. 143 (9), 2907-2916 (1989)
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  MEDLINE   90038522
FEATURES             Location/Qualifiers
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                     /db_xref="taxon:9606"
                     /cell_type="T-cell"
                     /germline
                     /map="17"
     mRNA            <1..520
                     /gene="SCYA1"
                     /note="G00-118-872"
     gene            1..520
                     /gene="SCYA1"
     CDS             51..341
                     /gene="SCYA1"
                     /codon_start=1
                     /db_xref="GDB:G00-118-872"
                     /product="secreted protein I-309"
                     /db_xref="PID:g339729"

/translation="MQIITTALVCLLLAGMWPEDVDSKSMQVPFSRCCFSFAEQEIPL
                     RAILCYRNTSSICSNEGLIFKLKRGKEACALDTVGWVQRHRKMLRHCPSKRK"
BASE COUNT      140 a    137 c    122 g    121 t
ORIGIN
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       61 tcaccacagc cctggtgtgc ttgctgctag ctgggatgtg gccggaagat gtggacagca
      121 agagcatgca ggtacccttc tccagatgtt gcttctcatt tgcggagcaa gagattcccc
      181 tgagggcaat cctgtgttac agaaatacca gctccatctg ctccaatgag ggcttaatat
      241 tcaagctgaa gagaggcaaa gaggcctgcg ccttggacac agttggatgg gttcagaggc
      301 acagaaaaat gctgaggcac tgcccgtcaa aaagaaaatg agcagatttc tttccattgt
      361 gggctctgga aaccacatgg cttcacctgt ccccgaaact accagcccta caccattcct
      421 tctgccctgc ttttgctagg tcacagagga tctgcttggt cttgataagc tatgttgttg
      481 cactttaaac atttaaatta tacaatcatc aacccccaac
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LOCUS       AB000887     687 bp    mRNA            PRI       05-JUN-1997
DEFINITION  Human mRNA for EBI1-ligand chemokine, complete cds.
ACCESSION   AB000887
NID         g2189952
KEYWORDS    EBI1-ligand chemokine; ELC.
SOURCE      Homo sapiens fetal tissue_lib:lung cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (bases 1 to 687)
  AUTHORS     Yoshida,R.,   Imai,T.,   Hieshima,K.,   Kusuda,J.,   Baba,M.,
Kitaura,M.,
            Nishimura,M., Kakizaki,M., Nomiyama,H. and Yoshie,O.
  TITLE     Direct Submission
  JOURNAL   Submitted (05-FEB-1997) to the DDBJ/EMBL/GenBank databases.
            Hisayuki   Nomiyama,   Kumamoto   University   Medical   School,
Department
            of Biochemistry; Honjo 2-2-1, Kumamoto, Kumamoto 860, Japan
            (E-mail:nomiyama@gpo.kumamoto-u.ac.jp, Tel:+81-96-373-5063)
REFERENCE   2  (sites)
  AUTHORS     Yoshida,R.,   Imai,T.,   Hieshima,K.,   Kusuda,J.,   Baba,M.,
Kitaura,M.,
            Nishimura,M., Kakizaki,M., Nomiyama,H. and Yoshie,O.
  TITLE     Molecular cloning of a novel human CC chemokine EBI1-ligand
            chemokine that is a specific functional ligand for EBI1, CCR7
  JOURNAL   J. Biol. Chem. 272 (21), 13803-13809 (1997)
  MEDLINE   97298088
FEATURES             Location/Qualifiers
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                     /db_xref="taxon:9606"
                     /dev_stage="fetal"
                     /tissue_lib="lung"
     gene            139..435
                     /gene="ELC"
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     CDS              139..435
                      /gene="ELC"
                      /note="CC chemokine"
                      /codon_start=1
                      /product="EBI1-ligand chemokine"
                      /db_xref="PID:d1021215"
                      /db_xref="PID:g2189953"

/translation="MALLLALSLLVLWTSPAPTLSGTNDAEDCCLSVTQKPIPGYIVR

NFHYLLIKDGCRVPAVVFTTLRGRQLCAPPDQPWVERIIQRLQRTSAKMKRRSS"
     mat_peptide      202..432
                      /gene="ELC"
                      /product="EBI1-ligand chemokine"
     polyA_signal     657..662
BASE COUNT      154 a    223 c    173 g    137 t
ORIGIN
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       61 agcctcacac agatcctgca cacacccaga cagctggcgc tcacacattc accgttggcc
      121 tgcctctgtt caccctccat ggccctgcta ctggccctca gcctgctggt tctctggact
      181 tccccagccc caactctgag tggcaccaat gatgctgaag actgctgcct gtctgtgacc
      241 cagaaaccca tccctgggta catcgtgagg aacttccact accttctcat caaggatggc
      301 tgcagggtgc ctgctgtagt gttcaccaca ctgaggggcc gccagctctg tgcaccccca
      361 gaccagccct gggtagaacg catcatccag agactgcaga ggacctcagc caagatgaag
      421 cgccgcagca gttaacctat gaccgtgcag agggagcccg gagtccgagt caagcattgt
      481 gaattattac ctaacctggg gaaccgagga ccagaaggaa ggaccaggct tccagctcct
      541 ctgcaccaga cctgaccagc caggacaggg cctggggtgt gtgtgagtgt gagtgtgagc
      601 gagagggtga gtgtggtcag agtaaagctg ctccaccccc agattgcaat gctaccaata
      661 aagccgcctg gtgtttacaa ctaattg
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LOCUS       AB000221       760 bp    mRNA            PRI       31-JUL-1997
DEFINITION  Homo sapiens mRNA for CC chemokine, complete cds.
ACCESSION   AB000221
NID         g2289718
KEYWORDS        CC  chemokine;  PARC;  pulmonary  and  activation-regulated
chemokine.
SOURCE      Homo sapiens lung cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (bases 1 to 760)
  AUTHORS   Nomiyama,H.
  TITLE     Direct Submission
  JOURNAL   Submitted (04-JAN-1997) to the DDBJ/EMBL/GenBank databases.
            Hisayuki  Nomiyama,  Kumamoto  University  Medical  School,
Department
            of Biochemistry; Honjo 2-2-1, Kumamoto, Kumamoto 860, Japan
            (E-mail:nomiyama@gpo.kumamoto-u.ac.jp, Tel:81-96-373-5063,
            Fax:81-96-372-6140)
REFERENCE   2  (sites)
  AUTHORS   Hieshima,K., Imai,T., Baba,M., Shoudai,K., Ishizuka,K.,
            Nakagawa,T., Tsuruta,J., Takeya,M., Sakaki,Y., Takatsuki,K.,
            Miura,R., Opdenakker,G., Damme,J., Yoshie,O. and Nomiyama,H.
  TITLE     A novel human CC chemokine PARC that is most homologous to
            macrophage-inflammatory     protein-1alpha/LD78alpha      and
chemotactic
            for T lymphocytes, but not for monocytes
  JOURNAL   J. Immunol. 159 (3), 1140-1149 (1997)
  MEDLINE   97376836
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     CDS             64..333
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                     VDYSETSPQCPKPGVILLTKRGRQICADPNKKWVQKYISDLKLNA"
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ORIGIN
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       61 atcatgaagg gccttgcagc tgccctcctt gtcctcgtct gcaccatggc cctctgctcc
      121 tgtgcacaag ttggtaccaa caaagagctc tgctgcctcg tctatacctc ctggcagatt
      181 ccacaaaagt tcatagttga ctattctgaa accagccccc agtgccccaa gccaggtgtc
      241 atcctcctaa ccaagagagg ccggcagatc tgtgctgacc ccaataagaa gtgggtccag
      301 aaatacatca gcgacctgaa gctgaatgcc tgaggggcct ggaagctgcg agggcccagt
      361 gaacttggtg ggcccaggag ggaacaggag cctgagccag ggcaatggcc ctgccaccct
      421 ggaggccacc tcttctaaga gtcccatctg ctatgcccag ccacattaac taactttaat
      481 cttagtttat gcatcatatt tcattttgaa attgatttct attgttgagc tgcattatga
      541 aattagtatt ttctctgaca tctcatgaca ttgtctttat catcctttcc cctttccctt
      601 caactcttcg tacattcaat gcatggatca atcagtgtga ttagctttct cagcagacat
      661 tgtgccatat gtatcaaatg acaaatcttt attgaatggt tttgctcagc accacctttt
      721 aatatattgg cagtacttat tatataaaag gtaaaccagc
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LOCUS       D86955       799 bp    mRNA          PRI       06-MAR-1997
DEFINITION  Human mRNA for CC chemokine LARC precursor, complete cds.
ACCESSION   D86955
NID         g1871138
KEYWORDS    CC chemokine LARC precursor.
SOURCE      Homo sapiens cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (sites)
  AUTHORS   Hieshima,K., Imai,T., Opdenakker,G., Van Damme,J., Kusuda,J.,
            Tei,H., Sakaki,Y., Takatsuki,K., Miura,R., Yoshie,O. and
            Nomiyama,H.
  TITLE     Molecular cloning of a novel human CC chemokine liver and
            activation-regulated chemokine (LARC) expressed in liver.
            Chemotactic activity for lymphocytes and gene localization on
            chromosome 2
  JOURNAL   J. Biol. Chem. 272 (9), 5846-5853 (1997)
  MEDLINE   97190319
REFERENCE   2  (bases 1 to 799)
  AUTHORS   Hieshima,K., Imai,T., Opdenakker,G., Van Damme,J., Kusuda,J.,
            Tei,H., Sakaki,Y., Takatsuki,K., Miura,R., Yoshie,O. and
            Nomiyama,H.
  JOURNAL   Unpublished (1996)
REFERENCE   3  (bases 1 to 799)
  AUTHORS   Nomiyama,H.
  TITLE     Direct Submission
  JOURNAL   Submitted (08-AUG-1996) to the DDBJ/EMBL/GenBank databases.
            Hisayuki   Nomiyama,   Kumamoto   University   Medical   School,
Department
            of Biochemistry; Honjo 2-2-1, Kumamoto, Kumamoto 860, Japan
            (E-mail:nomiyama@gpo.kumamoto-u.ac.jp, Tel:+81-96-373-5063)
FEATURES             Location/Qualifiers
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                     /db_xref="taxon:9606"
                     /chromosome="2"
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                     /db_xref="PID:d1013880"
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     gene            59..349
                     /gene="LARC"
     mat_peptide     137..346
                     /gene="LARC"
                     /product="CC chemokine LARC"
BASE COUNT      240 a    138 c    153 g    268 t
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      121 cggcgaatca gaagcagcaa gcaactttga ctgctgtctt ggatacacag accgtattct
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LOCUS       HUMAR          538 bp    mRNA            PRI       11-SEP-1996
DEFINITION  Human mRNA for chemokine, complete cds.
ACCESSION   D43767
NID         g1536878
KEYWORDS    chemokine, thymus and activation-regulated; chemokine.
SOURCE      Homo sapiens male peripheral blood cDNA to mRNA, clone:D3A.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (sites)
  AUTHORS   Imai,T., Yoshida,T., Baba,M., Nishimura,M., Kakizaki,M. and
            Yoshie,O.
  TITLE       Molecular  cloning  of  a  novel  T  cell-directed  CC  chemokine
expressed
            in  thymus  by  signal  sequence  trap  using  Epstein-Barr  virus
vector
  JOURNAL   J. Biol. Chem. 271 (35), 21514-21521 (1996)
  MEDLINE   96355526
REFERENCE   2  (bases 1 to 538)
  AUTHORS   Imai,T.
  JOURNAL   Unpublished (1996)
REFERENCE   3  (bases 1 to 538)
  AUTHORS   Imai,T.
  TITLE     Direct Submission
  JOURNAL     Submitted  (07-DEC-1994)  to  the  DDBJ/EMBL/GenBank  databases.
Toshio
            Imai, Shionogi Institute for Medical Science; 2-5-1 Mishima,
            Settsu, Osaka 566, Japan (Tel:06-382-2612, Fax:06-382-2598)
FEATURES             Location/Qualifiers
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      181 ccttagaaag ctgaagacgt ggtaccagac atctgaggac tgctccaggg atgccatcgt
      241 ttttgtaact gtgcagggca gggccatctg ttcggacccc aacaacaaga gagtgaagaa
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      361 actgtctccc gggactacct gggacctcca ccgttggtgt tcaccgcccc caccctgagc
      421 gcctgggtcc aggggaggcc ttccagggac gaagaagagc cacagtgagg gagatcccat
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LOCUS       HUMEOTAXIN    807 bp    mRNA            PRI       25-SEP-1996
DEFINITION  Human mRNA for eotaxin, complete cds.
ACCESSION   D49372
NID         g1552240
KEYWORDS    eotaxin; eosinophil-selective CC chemokine; chemoattractant.
SOURCE      Homo sapiens Small intestine, proximal cDNA to mRNA, clone:141.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;    Mammalia;    Eutheria;    Primates;    Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (bases 1 to 807)
  AUTHORS   Kitaura,M., Nakajima,T., Imai,T., Harada,S., Combadiere,C.,
            Tiffany,H.L., Murphy,P.M. and Yoshie,O.
  TITLE     Molecular cloning of human eotaxin, an eosinophil-selective CC
            chemokine, and identification of a specific eosinophil eotaxin
            receptor, CC chemokine receptor 3
  JOURNAL   J. Biol. Chem. 271 (13), 7725-7730 (1996)
  MEDLINE   96205964
REFERENCE   2  (bases 1 to 807)
  AUTHORS   Yoshie,O.
  TITLE     Direct Submission
  JOURNAL    Submitted (15-FEB-1995) to the DDBJ/EMBL/GenBank databases.
Osamu
            Yoshie, Shionogi Institute for Medical Science; 2-5-1 Mishima,
            Settsu, Osaka 566, Japan (E-mail:osamu.yoshie@shionogi.co.jp,
            Tel:06-382-2612, Fax:06-382-2598)
COMMENT     On Sep 20, 1996 this sequence version replaced gi:1313900.
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     polyA_signal    775..780
     polyA_site      807
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      121 tgtggctgct gctcatagca gctgccttca gcccccaggg gctcgctggg ccagcttctg
      181 tcccaaccac ctgctgcttt aacctggcca ataggaagat accccttcag cgactagaga
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      421 agcctgagtg ttgcctaatt tgttttccct tcttacaatg cattctgagg taacctcatt
      481 atcagtccaa agggcatggg ttttattata tatatatata tttttttttt aaaaaaaaac
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LOCUS       HSCCCHEM      232 bp    RNA             PRI       10-SEP-1996
DEFINITION  H.sapiens mRNA for CC-chemokine.
ACCESSION   Z69291
NID         g1181148
KEYWORDS    CC-chemokine.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 232)
  AUTHORS   Bartels,J.H., Schlueter,C., Richter,E., Christophers,E. and
            Schroeder,J.M.
  TITLE     Cloning of a novel human chemokine homologous to human monocyte
            chemoattractant proteins and rodent eotaxins
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 232)
  AUTHORS   Bartels,J.H.
  TITLE     Direct Submission
  JOURNAL   Submitted (01-FEB-1996) Bartels J. H.,
            Christian-Albrechts-Universitaet            zu            Kiel,
Dermatology/Hautklinik,
            Mol.Biol.Lab.609, Schittenhelmstr. 7, Kiel, Schleswig-Holstein,
            Germany, D-24105
REFERENCE   3  (bases 1 to 232)
  AUTHORS   Bartels,J., Schluter,C., Richter,E., Noso,N., Kulke,R.,
            Christophers,E. and Schroder,J.M.
  TITLE       Human dermal fibroblasts express eotaxin: molecular cloning,
mRNA
            expression, and identification of eotaxin sequence variants
  JOURNAL   Biochem. Biophys. Res. Commun. 225 (3), 1045-1051 (1996)
  MEDLINE   96374440
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MCP-3
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LOCUS       HSHCC1GEN    4037 bp    DNA             PRI       01-OCT-1995
DEFINITION  H.sapiens gene for chemokine HCC-1.
ACCESSION   Z49269
NID         g1004266
KEYWORDS    chemokine.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 4037)
  AUTHORS      Pardigol,A.,   Maegert,H.J.,   Cieslak,A.,   Hill,O.,   Schulz-
Knappe,P.
            and Forssmann,W.G.
  TITLE     Nucleotide Sequence of the Gene for the Human Chemokine HCC-1
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 4037)
  AUTHORS   Pardigol,A.
  TITLE     Direct Submission
  JOURNAL     Submitted (18-MAY-1995) Andreas Pardigol, Molecular Biology,
Lower
            Saxony Institute for Peptide Research, Feodor-Lynen-Strasse 31,
            Hannover, Lower Saxon, 30625, Germany
FEATURES             Location/Qualifiers
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                     /sex="male"
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                     /product="chemokine HCC-1"
                     /db_xref="PID:g1004267"

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     exon            3021..3135
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      121 tgattctaag cctgtttcct caactgctgt tctaaagatt aaataggcta atattcataa
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4021 aacagtagca gaccccg
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LOCUS       HSCC21        925 bp    mRNA            PRI       30-JUN-1998
DEFINITION  H.sapiens mRNA for chemokine CC-2 and CC-1.
ACCESSION   Z70292
NID         g1296608
KEYWORDS    chemokine CC-1; chemokine CC-2.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 925)
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  AUTHORS   Pardigol,A., Forssmann,U., Zucht,H.D., Loetscher,P.,
            Schulz-Knappe,P., Baggiolini,M., Forssmann,W.G. and Magert,H.J.
  TITLE      HCC-2, a human chemokine: gene structure, expression pattern,
 and
            biological activity
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 95 (11), 6308-6313 (1998)
  MEDLINE   98263352
REFERENCE   2  (bases 1 to 925)
  AUTHORS   Pardigol,A.
  TITLE     Direct Submission
  JOURNAL     Submitted (25-MAR-1996) Andreas Pardigol, IV - Molecular
Biology,
            Lower Saxony Institute for Peptide Research, Feodor-Lynen-
Strasse
            31, Hannover, Lower Saxony, 30625, Germany
FEATURES             Location/Qualifiers
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                     /tissue_type="liver"
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the
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      181 agttctgaac agctttcact ttgctgctga ctgctgcacc tcctacatct cacaaagcat
      241 cccgtgttca ctcatgaaaa gttattttga aacgagcagc gagtgctcca agccaggtgt
      301 catattcctc accaagaagg ggcggcaagt ctgtgccaaa cccagtggtc cgggagttca
      361 ggattgcatg aaaaagctga agccctactc aatataataa taaagagaca aaagaggcca
      421 gccacccacc tccaacacct cctgagcctc tgaagctccc accaggccag ctctcctccc
      481 acaacagctt cccacagcat gaagatctcc gtggctgcca ttcccttctt cctcctcatc
      541 accatcgccc tagggaccaa gactgaatcc tcctcacggg gaccttacca cccctcagag
      601 tgctgcttca cctacactac ctacaagatc ccgcgtcagc ggattatgga ttactatgag
      661 accaacagcc agtgctccaa gcccggaatt gtcttcatca ccaaaagggg ccattccgtc
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      781 gtgacccaga aggggtggcg aaggcacagc tcagagacat aaagagaaga tgccaaggcc
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LOCUS       HSCC23          973 bp    RNA             PRI       03-MAY-1996
DEFINITION  H.sapiens mRNA for chemokine CC-2 and CC-3.
ACCESSION   Z70293
NID         g1296611
KEYWORDS    Human chemokine CC-2; Human chemokine CC-3.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 973)
  AUTHORS   Pardigol,A., Maegert,H.J., Zucht,HD., Forssmann,W.G. and
            Schulz-Knappe,P.
  TITLE     Transcription of a Human Tandem Gene results in a Mature
            Bicistronic mRNA encoding two Novel CC-Chemokines
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 973)
  AUTHORS   Pardigol,A.
  TITLE     Direct Submission
  JOURNAL     Submitted (25-MAR-1996) Andreas Pardigol, IV - Molecular
Biology,
            Lower Saxony Institute for Peptide Research, Feodor-Lynen-
Strasse
            31, Hannover, Lower Saxony, 30625, Germany
FEATURES             Location/Qualifiers
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                     /db_xref="taxon:9606"
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                     /tissue_type="liver"
                     /clone_lib="PCR fragments"
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                     mRNA"
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     3'UTR           829..973
     polyA_signal    950..956
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       61 ggtctccgtg gctgccctct cctgcctcat gcttgttgct gtccttggat cccaggccca
      121 gttcacaaat gatgcagaga cagagttaat gatgtcaaag cttccactgg aaaatccagt
      181 agttctgaac agctttcact ttgctgctga ctgctgcacc tcctacatct cacaaagcat
      241 cccgtgttca ctcatgaaaa gttattttga aacgagcagc gagtgctcca agccaggtgt
      301 catattcctc accaagaagg ggcggcaagt ctgtgccaaa cccagtggtc cgggagttca
      361 ggattgcatg aaaaagctga agccctactc aatataataa taaagagaca aaagaggcca
      421 gccacccacc tccaacacct cctgagcctc tgaagctccc accaggccag ctctcctccc
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  AUTHORS     Pardigol,A., Forssmann,U., Zucht,H.D., Loetscher,P.,
              Schulz-Knappe,P., Baggiolini,M., Forssmann,W.G. and Magert,H.J.
  TITLE        HCC-2, a human chemokine: gene structure, expression pattern.
and
              biological activity
  JOURNAL     Proc. Natl. Acad. Sci. U.S.A. 95 (11), 6308-6313 (1998)
  MEDLINE     98263352
REFERENCE     2  (bases 1 to 925)
  AUTHORS     Pardigol,A.
  TITLE       Direct Submission
  JOURNAL       Submitted (25-MAR-1996) Andreas Pardigol, IV - Molecular
Biology,
              Lower Saxony Institute for Peptide Research, Feodor-Lynen-
Strasse
              31, Hannover, Lower Saxony, 30625, Germany
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                       /db_xref="taxon:9606"
                       /dev_stage="adult"
                       /tissue_type="liver"
                       /clone_lib="PCR fragments"
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     CDS               56..397
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                       /db_xref="PID:g1296609"
                       /db_xref="SWISS-PROT:Q16663"

/translation="MKVSVAALSCLMLVAVLGSQAQFTNDAETELMMSKLPLENPVVL

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     misc_feature      398..498
                       /note="spacing region between two coding regions of
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                       bicistronic mRNA"
     CDS               499..780
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                       /db_xref="PID:g1296610"
                       /db_xref="SWISS-PROT:Q16627"

/translation="MKISVAAIPFFLLITIALGTKTESSSRGPYHPSECCFTYTTYKI
                       PRQRIMDYYETNSQCSKPGIVFITKRGHSVCTNPSDKWVQDYIKDMKEN"
     3'UTR             781..925
     polyA_signal      902..908
BASE COUNT      240 a    296 c    199 g    190 t
ORIGIN
        1 ccaggaagca gtgagcccag gagtcctcgg ccagccctgc ctgcccacca ggaggatgaa
       61 ggtctccgtg gctgccctct cctgcctcat gcttgttgct gtccttggat cccaggccca
      121 gttcacaaat gatgcagaga cagagttaat gatgtcaaag cttccactgg aaaatccagt
      181 agttctgaac agctttcact ttgctgctga ctgctgcacc tcctacatct cacaaagcat
      241 cccgtgttca ctcatgaaaa gttattttga aacgagcagc gagtgctcca agccaggtgt
      301 catattcctc accaagaagg ggcggcaagt ctgtgccaaa cccagtggtc cgggagttca
      361 ggattgcatg aaaaagctga agccctactc aatataataa taaagagaca aaagaggcca
      421 gccacccacc tccaacacct cctgagcctc tgaagctccc accaggccag ctctcctccc
      481 acaacagctt cccacagcat gaagatctcc gtggctgcca ttcccttctt cctcctcatc
      541 accatcgccc tagggaccaa gactgaatcc tcctcacggg gaccttacca cccctcagag
      601 tgctgcttca cctacactac ctacaagatc ccgcgtcagc ggattatgga ttactatgag
      661 accaacagcc agtgctccaa gcccggaatt gtcttcatca ccaaaagggg ccattccgtc
      721 tgtaccaacc ccagtgacaa gtgggtccag gactatatca aggacatgaa ggagaactga
      781 gtgacccaga aggggtggcg aaggcacagc tcagagacat aaagagaaga tgccaaggcc
      841 ccctcctcca cccaccgcta actctcagcc ccagtcaccc tcttggagct tccctgcttt
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     1141 tttttcatag gaagtccgga tgggaatatt cacattaatc atttttgcag agactttgct
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     1261 aaaaaaaaaa aaagagagag agagagaaga agaagaagaa gagacacaaa tctctacctc
     1321 ccatgttaag ctttgcagga cagggaaaga aagggtatga gacacggcta ggggtaaact
     1381 cttagtccaa aacccaagca tgcaataaat aaaactccct tatttgacaa
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LOCUS       AB007454     1503 bp    mRNA            PRI       09-APR-1998
DEFINITION  Homo sapiens mRNA for chemokine LEC precursor, complete cds.
ACCESSION   AB007454
NID         g2723285
KEYWORDS    chemokine LEC precursor.
SOURCE      Homo sapiens liver cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (sites)
  AUTHORS   Shoudai,K., Hieshima,K., Fukuda,S., Iio,M., Miura,R., Imai,T.,
            Yoshie,O. and Nomiyama,H.
  TITLE     Isolation of cDNA encoding a novel human CC chemokine NCC-4/LEC
  JOURNAL   Biochim. Biophys. Acta 1396 (3), 273-277 (1998)
  MEDLINE   98207719
REFERENCE   2  (bases 1 to 1503)
  AUTHORS   Nomiyama,H.
  TITLE     Direct Submission
  JOURNAL   Submitted (19-SEP-1997) to the DDBJ/EMBL/GenBank databases.
            Hisayuki  Nomiyama,  Kumamoto  University  Medical  School,
Department
            of  Biochemistry;  Honjo  2-2-1,  Kumamoto,  Kumamoto  860-0811,
Japan
            (E-mail:nomiyama@gpo.kumamoto-u.ac.jp, Tel:81-96-373-5063,
            Fax:81-96-372-6140)
FEATURES             Location/Qualifiers
     source          1..1503
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     CDS             77..439
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                     /db_xref="PID:d1024963"
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/translation="MKVSEAALSLLVLILIITSASRSQPKVPEWVNTPSTCCLKYYEK

VLPRRLVVGYRKALNCHLPAIIFVTKRNREVCTNPNDDWVQEYIKDPNLPLLPTRNLS
                     TVKIITAKNGQPQLLNSQ"
     mat_peptide     146..436
     polyA_signal    560..565
     polyA_signal    1485..1490
BASE COUNT      417 a    374 c    312 g    400 t
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       61 ctgcctcgct gagaggatga aggtctccga ggctgccctg tctctccttg tcctcatcct
      121 tatcattact tcggcttctc gcagccagcc aaaagttcct gagtgggtga acaccccatc
      181 cacctgctgc ctgaagtatt atgagaaagt gttgccaagg agactagtgg tgggatacag
      241 aaaggccctc aactgtcacc tgccagcaat catcttcgtc accaagagga accgagaagt
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      421 gctcctcaac tcccagtgat gaccaggctt tagtggaagc ccttgtttac agaagagagg
      481 ggtaaaccta tgaaaacagg ggaagcctta ttaggctgaa actagccagt cacattgaga
      541 gaagcagaac aatgatcaaa ataaaggaga agtatttcga atattttctc aatcttagga
      601 ggaaatacca aagttaaggg acgtgggcag aggtacgctc ttttattttt atatttatat
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LOCUS       HSU91746     1430 bp    mRNA            PRI       12-MAR-1998
DEFINITION  Homo sapiens IL-10-inducible chemokine (HCC-4) mRNA, complete
cds.
ACCESSION   U91746
NID         g2581780
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1430)
  AUTHORS   Hedrick,J.A., Helms,A., Gorman,D. and Zlotnik,A.
  TITLE     Identification of a novel human CC chemokine upregulated by IL-
10
  JOURNAL   Blood (1998) In press
REFERENCE   2  (bases 1 to 1430)
  AUTHORS   Hedrick,J.A., Helms,A., Gorman,D. and Zlotnik,A.
  TITLE     Direct Submission
  JOURNAL    Submitted (02-MAR-1997) Immunology, DNAX Research Institute,
901
            California Ave, Palo Alto, CA 94304, USA
FEATURES             Location/Qualifiers
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                     /db_xref="PID:g2581781"

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      661 tggctcactg taacctccac ctcccaggct caagtgatcc tcccacccca gcctcccgag
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     1021 ttgccctctg ggtcttgacc cattaagagc cccatagaac tcttgatttt tcctgtccat
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                Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
                Homo.
REFERENCE       1  (bases 1 to 821)
  AUTHORS       Hromas,R., Gray,P.W., Chantry,D., Godiska,R., Krathwohl,M.,
                Fife,K.,   Bell,G.I.,   Takeda,J.,   Aronica,S.,   Gordon,M.,
Cooper,S.,
                Broxmeyer,H.E. and Klemsz,M.J.
  TITLE         Cloning and characterization of exodus, a novel beta-chemokine
  JOURNAL       Blood 89 (9), 3315-3322 (1997)
  MEDLINE       97275143
REFERENCE       2  (bases 1 to 821)
  AUTHORS       Hromas,R.A.
  TITLE         Direct Submission
  JOURNAL       Submitted (17-JUL-1996) Indiana University Medical Center,
                Medicine, 975 W. Walnut St., Indianapolis, IN 46202, USA
FEATURES                 Location/Qualifiers
     source              1..821
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                         progenitors and HIV"
                         /codon_start=1
                         /product="chemokine exodus-1"
                         /db_xref="PID:g1778717"
/translation="MCCTKSLLLAALMSVLLLHLCGESEASNFDCCLGYTDRILHPKF
                         IVGFTRQLANEGCDINAIIFHTKKKLSVCANPKQTWVKYIVRLLSKKVKNM"
     variation           121^122
                         /note="insertion of an extra codon GCA at nt 121,
encoding
                         for an alanine after the alanine at amino acid
position
                         26, represents the allelic difference of the
transcript
                         isolated from macrophages"
BASE COUNT      258 a    134 c    156 g    273 t
ORIGIN
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LOCUS       HSU88320     828 bp    mRNA            PRI       18-DEC-1997
DEFINITION  Human beta chemokine Exodus-2 mRNA, complete cds.
ACCESSION   U88320
NID         g2196919
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 828)
  AUTHORS      Hromas,R.,  Kim,C.H.,  Klemsz,M.,  Krathwohl,M.,  Fife,K.,
Cooper,S.,
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          Schnizlein-Bick,C. and Broxmeyer,H.E.
  TITLE      Isolation and characterization of Exodus-2, a novel C-C
chemokine
          with a unique 37-amino acid carboxyl-terminal extension
  JOURNAL   J. Immunol. 159 (6), 2554-2558 (1997)
  MEDLINE   97444139
REFERENCE   2  (bases 1 to 828)
  AUTHORS   Hromas,R.A.
  TITLE     Direct Submission
  JOURNAL   Submitted (04-FEB-1997) Medicine, Indiana University Medical
            Center, 975 West Walnut, Indianapolis, IN 46202, USA
FEATURES             Location/Qualifiers
     source          1..828
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                     /note="PCR amplified from activated THP-1 cells"
                     /db_xref="taxon:9606"
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                     /cell_line="THP-1"
                     /cell_type="monoblast"
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                     /product="beta chemokine Exodus-2"
                     /db_xref="PID:g2196920"

/translation="MAQSLALSLLILVLAFGIPRTQGSDGGAQDCCLKYSQRKIPAKV

VRSYRKQEPSLGCSIPAILFLPRKRSQAELCADPKELWVQQLMQHLDKTPSPQKPAQG
                     CRKDRGASKTGKKGKGSKGCKRTERSQTPKGP"
BASE COUNT      218 a    255 c    216 g    139 t
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       61 ttggcatccc caggacccaa ggcagtgatg gaggggctca ggactgttgc ctcaagtaca
      121 gccaaaggaa gattcccgcc aaggttgtcc gcagctaccg gaagcaggaa ccaagcttag
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      661 ggagcatgag aggaggcagc aggactgtcc ccttgaagga gaatcatcag gaccctggac
      721 ctgatacggc tccccagtac accccacctc ttccttgtaa atatgattta tacctaactg
      781 aataaaaagc tgttctgtct tcccacccaa aaaaaaaaa aaaaaaa
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LOCUS       HSU88321     502 bp    mRNA            PRI       22-JUN-1998
DEFINITION  Human beta chemokine Exodus-3 mRNA, complete cds.
ACCESSION   U88321
NID         g2196921
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 502)
  AUTHORS   Hromas,R.A., Gray,P., Klemsz,M., Fife,K. and Broxmeyer,H.
  TITLE     DCCL chemokines represent a novel beta chemokine subfamily
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 502)
  AUTHORS   Hromas,R.A.
  TITLE     Direct Submission
  JOURNAL   Submitted (04-FEB-1997) Medicine, Indiana University Medical
            Center, 975 West Walnut, Indianapolis, IN 46202, USA
REFERENCE   3  (bases 1 to 502)
  AUTHORS   Hromas,R.A.
  TITLE     Direct Submission
  JOURNAL   Submitted (22-JUN-1998) Medicine, Indiana University Medical
            Center, 975 West Walnut, Indianapolis, IN 46202, USA
  REMARK    Amino acid sequence updated by submitter
FEATURES             Location/Qualifiers
     source          1..502
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                     /organism="Homo sapiens"
                     /note="PCR amplified from THP-1 cells"
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                     /dev_stage="adult"
     CDS             120..416
                     /note="Mip-3alpha/ELC/CKbeta11"
                     /codon_start=1
                     /product="beta chemokine Exodus-3"
                     /db_xref="PID:g3243080"

/translation="MALLLALSLLVLWTSPAPTLSGTNDAEDCCLSVTQKPIPGYIVR

NFHYLLIKDGCRVPAVVFTTLRGRQLCAPPDQPWVERIIQRLQRTSAKMKRRSS"
BASE COUNT      113 a    170 c    121 g     98 t
ORIGIN
        1 ctcacacctt gcatttcacc cctgcatccc atgcgccctg cagcctcaca cagatcctgc
       61 acacacccag acagctggcg ctcacacatt caccgttggc ctgcctctgt tcaccctcca
      121 tggccctgct actggccctc agcctgctgg ttctctggac ttccccagcc ccaactctga
      181 gtggcaccaa tgatgctgaa gactgctgcc tgtctgtgac ccagaaaccc atccctgggt
      241 acatcgtgag gaacttccac taccttctca tcaaggatgg ctgcagggtg cctgctgtag
      301 tgttcaccac actgaggggc cgccagctct gtgcaccccc agaccagccc tgggtagaac
      361 gcatcatcca gagactgcag aggacctcag ccaagatgaa gcgccgcagc agttaaccta
      421 tgaccgtgca gagggagccc cgagtccgag tcaagcattg tgaattatta ctaactggga
      481 acgaggacag aaggaaggac ag
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LOCUS       HSU86358      879 bp    mRNA            PRI       11-SEP-1997
DEFINITION  Human chemokine (TECK) mRNA, complete cds.
ACCESSION   U86358
NID         g2388626
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 879)
  AUTHORS       Vicari,A.P.,   Figueroa,D.J.,   Hedrick,J.A.,   Foster,J.S.,
Singh,K.P.,
            Menon,S., Copeland,N.G., Gilbert,D.J., Jenkins,N.A., Bacon,K.B.
and
            Zlotnik,A.
  TITLE     TECK: a novel cc chemokine specifically expressed by thymic
            dendritic cells and potentially involved in T cell development
  JOURNAL   Immunology 7, 291-301 (1997)
REFERENCE   2  (bases 1 to 879)
  AUTHORS   Vicari,A.P. and Zlotnik,A.
  TITLE     Direct Submission
  JOURNAL     Submitted (21-JAN-1997) Immunology, DNAX Research Institute,
901
            California Ave., Palo Alto, CA 94304, USA
FEATURES             Location/Qualifiers
     source          1..879
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                     /db_xref="taxon:9606"
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     CDS             1..453
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                     /product="chemokine"
                     /db_xref="PID:g2388627"

/translation="MNLWLLACLVAGFLGAWAPAVHTQGVFEDCCLAYHYPIGWAVLR

RAWTYRIQEVSGSCNLPAAIFYLPKRHRKVCGNPKSREVQRAMKLLDARNKVFAKLHH
                     NMQTFQAGPHAVKKLSSGNSKLSSSKFSNPISSSKRNVSLLISANSGL"
BASE COUNT      191 a    264 c    218 g    206 t
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ORIGIN
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      121 gctgtgctcc ggcgcgcctg gacttaccgg atccaggagg tgagcgggag ctgcaatctg
      181 cctgctgcga tattctacct ccccaagaga cacaggaagg tgtgtgggaa ccccaaaagc
      241 agggaggtgc agagagccat gaagctcctg gatgctcgaa ataaggtttt tgcaaagctc
      301 caccacaaca tgcagacctt ccaagcaggc cctcatgctg taaagaagtt gagttctgga
      361 aactccaagt tatcatcatc caagtttagc aatcccatca gcagcagcaa gaggaatgtc
      421 tccctcctga tatcagctaa ttcaggactg tgagccggct catttctggg ctccatcggc
      481 acaggagggg ccggatcttt ctccgataaa accgtcgccc tacagaccca gctgtcccca
      541 cgcctctgtc ttttgggtca agtcttaatc cctgcacctg agttggtcct ccctctgcac
      601 ccccaccacc tcctgcccgt ctggcaactg gaaagaagga gttggcctga ttttaacctt
      661 ttgccgctcc ggggaacagc acaatcctgg gcagccagtg gctcttgtag agaaaactta
      721 ggatacctct ctcactttct gtttcttgcc gtccaccccg ggccatgcca gtgtgtcctc
      781 tgggtcccct ccaaaaatct ggtcattcaa ggatcccctc ccaaggctat gcttttctat
      841 aacttttaaa taaaccttgg ggggtgaatg gaataaaaa
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LOCUS       AB002409       852 bp    mRNA            PRI       15-AUG-1997
DEFINITION  Homo sapiens mRNA for SLC, complete cds.
ACCESSION   AB002409
NID         g2335034
KEYWORDS    SLC; mature ELC.
SOURCE      Homo sapiens cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (bases 1 to 852)
  AUTHORS   Nomiyama,H.
  TITLE     Direct Submission
  JOURNAL   Submitted (28-MAR-1997) to the DDBJ/EMBL/GenBank databases.
            Hisayuki   Nomiyama,   Kumamoto   University   Medical   School,
Department
            of Biochemistry; Honjo 2-2-1, Kumamoto, Kumamoto 860, Japan
            (E-mail:nomiyama@gpo.kumamoto-u.ac.jp, Tel:81-96-373-5063,
            Fax:81-96-372-6140)
REFERENCE   2  (bases 1 to 852)
  AUTHORS      Nagira,M.,   Imai,T.,   Hieshima,K.,   Kusuda,J.,   Ridanpaa,M.,
Takagi,S.,
            Nishimura,M., Kakizaki,M., Nomiyama,H. and Yoshie,O.
  TITLE     Molecular Cloning of a Novel Human CC Chemokine Secondary
            Lymphoid-Tissue Chemokine (SLC) That is an Efficient
            Chemoattractant for Lymphocytes and Mapped to Chromosome 9p13
  JOURNAL   Unpublished (1997)
FEATURES             Location/Qualifiers
     source          1..852
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
     CDS             59..463
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                     /db_xref="PID:d1022673"
                     /db_xref="PID:g2335035"

/translation="MAQSLALSLLILVLAFGIPRTQGSDGGAQDCCLKYSQRKIPAKV

VRSYRKQEPSLGCSIPAILFLPRKRSQAELCADPKELWVQQLMQHLDKTPSPQKPAQG
                     CRKDRGASKTGKKGKGSKGCKRTERSQTPKGP"
     mat_peptide     <107..460
                     /product="mature ELC"
     polyA_site      823..828
BASE COUNT      205 a    279 c    217 g    151 t
ORIGIN
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       61 ggctcagtca ctggctctga gcctccttat cctggttctg gcctttggca tccccaggac
      121 ccaaggcagt gatggagggg ctcaggactg ttgcctcaag tacagccaaa ggaagattcc
      181 cgccaaggtt gtccgcagct accggaagca ggaaccaagc ttaggctgct ccatcccagc
      241 tatcctgttc ttgccccgca agcgctctca ggcagagcta tgtgcagacc caaaggagct
      301 ctgggtgcag cagctgatgc agcatctgga caagacacca tccccacaga aaccagccca
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      361 gggctgcagg aaggacaggg gggcctccaa gactggcaag aaaggaaagg gctccaaagg
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      481 agccctggag accccaccag cctcaccaac gcttgaagcc tgaacccaag atgcaagaag
      541 gaggctatgc tcaggggccc tggagcagcc accccatgct ggccttgcca cactctttct
      601 cctgctttaa ccaccccatc tgcattccca gctctaccct gcatggctga gctgcccaca
      661 gcaggccagg tccagagaga ccgaggaggg agagtctccc agggagcatg agaggaggca
      721 gcaggactgt ccccttgaag gagaatcatc aggaccctgg acctgatacg gctccccagt
      781 acaccccacc tcttccttgt aaatatgatt tatacctaac tgaataaaaa gctgttctgt
      841 cttcccaccc gc
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LOCUS       AF055467     1481 bp    mRNA            PRI       06-AUG-1998
DEFINITION  Homo sapiens monotactin-1 mRNA, complete cds.
ACCESSION   AF055467
NID         g3395775
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1481)
  AUTHORS   Youn,B.S., Zhang,S., Broxmeyer,H.E., Antol,K., Fraser,M.J. Jr.,
            Hangoc,G. and Kwon,B.S.
  TITLE     Isolation and characterization of LMC, a novel lymphocyte and
            monocyte    chemoattractant    human    CC    chemokine,    with
myelosuppressive
            activity
  JOURNAL   Biochem. Biophys. Res. Commun. 247 (2), 217-222 (1998)
  MEDLINE   98308096
REFERENCE   2  (bases 1 to 1481)
  AUTHORS   Youn,B.S. and Kwon,B.S.
  TITLE     Direct Submission
  JOURNAL   Submitted (24-MAR-1998) Microbiology and Immunology, Indiana
            University,  School  of  Medicine,  605  Barnhill  Dr.  Medical
Science
            Bldg., Indianapolis, IN 46202, USA
FEATURES             Location/Qualifiers
     source          1..1481
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                     /db_xref="taxon:9606"
                     /chromosome="17"
     5'UTR           1..34
     CDS             35..397
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                     CC chemokine"
                     /codon_start=1
                     /product="monotactin-1"
                     /db_xref="PID:g3395776"

/translation="MKVSEAALSLLVLILIITSASRSQPKVPEWVNTPSTCCLKYYEK

VLPRRLVVGYRKALNCHLPAIIFVTKRNREVCTNPNDDWVQEYIKDPNLPLLPTRNLS
                     TVKIITAKNGQPQLLNSQ"
     3'UTR           398..1481
BASE COUNT      412 a    362 c    302 g    405 t
ORIGIN
        1 gcacgagctg aagctgtact gcctcgctga gaggatgaag gtctccgagg ctgccctgtc
       61 tctccttgtc ctcatcctta tcattacttc ggcttctcgc agccagccaa aagttcctga
      121 gtgggtgaac accccatcca cctgctgcct gaagtattat gagaaagtgt tgccaaggag
      181 actagtggtg ggatacagaa aggccctcaa ctgtcacctg ccagcaatca tcttcgtcac
      241 caagaggaac cgagaagtct gcaccaaccc caatgacgac tgggtccaag agtacatcaa
      301 ggatcccaac ctacctttgc tgcctaccag gaacttgtcc acggttaaaa ttattacagc
      361 aaagaatggt caaccccagc tcctcaactc ccagtgatga ccaagcttta gtggaagccc
      421 ttgtttacag aagagagggg taaactatga aaacaggga agccttatta ggctgaaact
      481 agccagtcac attgagagaa gcagaacaat gatcaaaata aaggagaagt atttcgaata
      541 ttttctcaat cttaggagga aataccaaag ttaagggacg tgggcagagg tacgctcttt
      601 tatttttata tttatatttt tattttttg agatagggtc ttactctgtc acccaggctg
      661 gagtgcagtg gtgtgatctt ggctcacttg atcttggctc actgtaacct ccacctccca
      721 ggctcaagtg atcctcccac cccaccctcc cgagtagctg ggactacagg cttgcgccac
      781 cacacctggc taatttttgt atttttggta gagacgggat tctaccatgt tgcccaggct
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     841 ggtctcaaac tcgtgtgccc aagcaatcca cctgcctcag ccttccaaaa gtgctgggct
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     961 acatccttct cctgctctct ttcaattcct cacttcacac cagtacacaa gccattctaa
    1021 atacttagcc agtttccagc cttccagatg atctttgccc tctgggtctt gacccattaa
    1081 gagccccata gaactcttga tttttcctgt ccatctttat gggatttttc tggatctata
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    1201 atattcacat taatcatttt tgcagagact ttgctagatc ctctcatatt ttgtcttcct
    1261 cagggtggca ggggtacaga agtgcctgat tggttttttt tttttttgag agagagagag
    1321 aagaagaaga agaagagaca caaatctcta cctcccatgt taagctttgc aggacaggga
    1381 aagaaagggt atgagacacg gctagggtaa actcttagtc caaaacccaa gcatgcaata
    1441 aataaaactc ccttatttga caaaaaaaaa aaaaaaaaaa a
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LOCUS       HSRNAATAC     557 bp    RNA             PRI       06-JUL-1995
DEFINITION  H.sapiens mRNA for ATAC protein.
ACCESSION   X86474
NID         g895846
KEYWORDS    ATAC gene.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 557)
  AUTHORS   Muller,S., Dorner,B., Korthauer,U., Mages,H.W., D'Apuzzo,M.,
            Senger,G. and Kroczek,R.A.
  TITLE        Cloning  of  ATAC,  an  activation-induced,  chemokine-related
molecule
            exclusively expressed in CD8+ T lymphocytes
  JOURNAL   Eur. J. Immunol. 25 (6), 1744-1748 (1995)
  MEDLINE   95339892
REFERENCE   2  (bases 1 to 557)
  AUTHORS   Kroczek,R.A.
  TITLE     Direct Submission
  JOURNAL   Submitted (20-APR-1995) R.A. Kroczek, Molecular Immunology,
            Robert-Koch-Institute, Nordufer 20, 13353 Berlin, FRG
FEATURES             Location/Qualifiers
     source          1..557
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
                     /tissue_type="peripheral blood"
                     /cell_type="lymphocyte"
                     /chromosome="1"
                     /map="q23"
     gene            25..369
                     /gene="ATAC"
     CDS             25..369
                     /gene="ATAC"
                     /codon_start=1
                     /product="CD8+T cell specific protein"
                     /db_xref="PID:g895847"
                     /db_xref="SWISS-PROT:P47992"

/translation="MRLLILALLGICSLTAYIVEGVGSEVSDKRTCVSLTTQRLPVSR

IKTYTITEGSLRAVIFITKRGLKVCADPQATWVRDVVRSMDRKSNTRNNMIQTKPTGT
                     QQSTNTAVTLTG"
     polyA_signal    469..474
     polyA_signal    534..539
BASE COUNT      157 a    139 c    112 g    149 t
ORIGIN
        1 gcacagctca gcaggacctc agccatgaga cttctcatcc tggccctcct tggcatctgc
       61 tctctcactg catacattgt ggaaggtgta gggagtgaag tctcagataa gaggacctgt
      121 gtgagcctca ctacccagcg actgccggtt agcagaatca agacctacac catcacggaa
      181 ggctccttga gagcagtaat ttttattacc aaacgtggcc taaaagtctg tgctgatcca
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      361 actggctagt agtctctggc accctgtccg tctccagcca gccagctcat ttcactttac
      421 acgctcatgg actgagttta tactcgcctt ttatgaaagc actgcatgaa taaaattatt
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      541 tatttattat taagaat
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LOCUS       HSU85767      563 bp    mRNA            PRI       01-APR-1997
DEFINITION   Human  myeloid  progenitor  inhibitory  factor-1  MPIF-1  mRNA,
complete
            cds.
ACCESSION   U85767
NID         g1916249
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 563)
  AUTHORS   Patel,V.P., Kreider,B.L., Li,Y., Li,H., Leung,K., Salcedo,T.,
            Nardelli,B., Pippalla,V., Gentz,S., Thotakura,R., Parmelee,D.,
            Gentz,R. and Garotta,G.
  TITLE     Molecular and functional characterization of two novel human C-
C
            chemokines as inhibitors of two distinct classes of myeloid
            progenitors
  JOURNAL   J. Exp. Med. (1997) In press
REFERENCE   2  (bases 1 to 563)
  AUTHORS   Li,H. and Patel,V.P.
  TITLE     Direct Submission
  JOURNAL    Submitted (17-JAN-1997) Cell Biology, Human Genome Sciences,
9410
            Keywest Ave., Rockville, MD 20850, USA
FEATURES             Location/Qualifiers
     source          1..563
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                     /db_xref="taxon:9606"
     CDS             31..393
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                     /codon_start=1
                     /product="MPIF-1"
                     /db_xref="PID:g1916250"

/translation="MKVSVAALSCLMLVTALGSQARVTKDAETEFMMSKLPLENPVLL

DRFHATSADCCISYTPRSIPCSLLESYFETNSECSKPGVIFLTKKGRRFCANPSDKQV
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BASE COUNT      164 a    143 c    117 g    139 t
ORIGIN
        1 ctcagccagc cctgcctgcc caccaggagg atgaaggtct ccgtggctgc cctctcctgc
       61 ctcatgcttg ttactgccct tggatcccag gcccgggtca caaaagatgc agagacagag
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      181 agtgctgact gctgcatctc ctacacccca cgaagcatcc cgtgttcact cctggagagt
      241 tactttgaaa cgaacagcga gtgctccaag ccgggtgtca tcttcctcac caagaagggg
      301 cgacgtttct gtgccaaccc cagtgataag caagttcagg tttgcatgag aatgctgaag
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      421 gccagccacc aactttcttg cctcaactac cttcctgaat tattttttta agaagcattt
      481 attcttgtgt tctggattta gagcaattca tctaataaac agtttctcac ttttaaaaaa
      541 aaaaaaaaaa aaaaaaaaaa aaa
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LOCUS       HSU85768      360 bp    mRNA            PRI       01-APR-1997
DEFINITION   Human  myeloid  progenitor  inhibitory  factor-1  MPIF-2  mRNA,
complete
            cds.
ACCESSION   U85768
NID         g1916251
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 360)
  AUTHORS   Patel,V.P., Kreider,B.L., Li,Y., Li,H., Leung,K., Salcedo,T.,
            Nardelli,B., Pippalla,V., Gentz,S., Thotakura,R., Parmelee,D.,
            Gentz,R. and Garotta,G.
  TITLE     Molecular and functional characterization of two novel human C-
```

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C
            chemokines as inhibitors of two distinct classes of myeloid
            progenitors
  JOURNAL   J. Exp. Med. (1997) In press
REFERENCE   2  (bases 1 to 360)
  AUTHORS   Li,H. and Patel,V.P.
  TITLE     Direct Submission
  JOURNAL    Submitted (17-JAN-1997) Cell Biology, Human Genome Sciences,
9410
            Keywest Ave., Rockville, MD 20850, USA
FEATURES             Location/Qualifiers
     source          1..360
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
     CDS             1..360
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                     /db_xref="PID:g1916252"

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NRVVSYQLSSRSTCLKGGVIFTTKKGQQFCGDPKQEWVQRYMKNLDAKQKKASPRARA
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BASE COUNT       85 a    106 c     96 g     73 t
ORIGIN
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       61 atcatcccta cgggctctgt ggtcataccc tctccctgct gcatgttctt tgtttccaag
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      241 tgggtccaga ggtacatgaa gaacctggac gccaagcaga agaaggcttc ccctagggcc
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LOCUS       HUMSDF1A      1847 bp    mRNA            PRI       26-DEC-1996
DEFINITION  Human pre-B cell stimulating factor homologue (SDF1a) mRNA,
            complete cds.
ACCESSION   L36034
NID         g1220363
KEYWORDS      intercrine; intercrine CXC subfamily; pre-B cell stimulating
factor
            homologue; alpha-chemokine.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1847)
  AUTHORS   Shirozu,M., Nakano,T., Inazawa,J., Tashiro,K., Tada,H.,
            Shinohara,T. and Honjo,T.
  TITLE     Structure and chromosomal localization of the human stromal
            cell-derived factor 1 (SDF1) gene
  JOURNAL   Genomics 28 (3), 495-500 (1995)
  MEDLINE   96039262
FEATURES             Location/Qualifiers
     source          1..1847
                     /organism="Homo sapiens"
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     CDS             80..349
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      121 cgcgctctgc ctcagcgacg ggaagcccgt cagcctgagc tacagatgcc catgccgatt
      181 cttcgaaagc catgttgcca gagccaacgt caagcatctc aaaattctca acactccaaa
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      301 gaagctaaag tggattcagg agtacctgga gaaagcttta aacaagtaag cacaacagcc
      361 aaaaaggact ttccgctaga cccactcgag gaaaactaaa accttgtgag agatgaaagg
      421 gcaaagacgt gggggagggg gccttaacca tgaggaccag gtgtgtgtgt ggggtgggca
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      541 gtatgatatt gcagcttata ttcatccatg ccctgtacct gtgcacgttg gaacttttat
      601 tactggggtt tttctaagaa agaaattgta ttatcaacag cattttcaag cagttagttc
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      781 agtcaggtgg tggcttaaca gggagctgga aaaagtgtcc tttcttcaga cactgaggct
      841 cccgcagcag cgcccctccc aagaggaagg cctctgtggc actcagatac cgactggggc
      901 tggggcgccg ccactgcctt cacctcctct ttcaaacctc agtgattggc tctgtgggct
      961 ccatgtagaa gccactatta ctgggactgt ctcagagacc cctctcccag ctattcctac
     1021 tctctccccg actccgagag catgcttaat cttgcttctg cttctcattt ctgtagcctg
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LOCUS       HUMSDF1B     3524 bp    mRNA            PRI       26-DEC-1996
DEFINITION  Human pre-B cell stimulating factor homologue (SDF1b) mRNA,
            complete cds.
ACCESSION   L36033
NID         g1220365
KEYWORDS     intercrine; intercrine CXC subfamily; pre-B cell stimulating
factor
            homologue; alpha-chemokine.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 3524)
  AUTHORS   Shirozu,M., Nakano,T., Inazawa,J., Tashiro,K., Tada,H.,
            Shinohara,T. and Honjo,T.
  TITLE     Structure and chromosomal localization of the human stromal
            cell-derived factor 1 (SDF1) gene
  JOURNAL   Genomics 28 (3), 495-500 (1995)
  MEDLINE   96039262
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LOCUS       HSJ002211     663 bp    mRNA            PRI       11-MAR-1998
DEFINITION  Homo sapiens cDNA for a CXC chemokine.
ACCESSION   AJ002211
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NID         g2832410
KEYWORDS    CXC chemokine.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 663)
  AUTHORS      Legler,D.F.,   Loetscher,M.,   Roos,R.S.,   Clark-Lewis,I.,
Baggiolini,M.
            and Moser,B.
  TITLE      B cell-attracting chemokine 1, a human CXC chemokine expressed
in
            lymphoid  tissues,  selectively  attracts  B  lymphocytes  via
BLR1/CXCR5
  JOURNAL   J. Exp. Med. 187 (4), 655-660 (1998)
  MEDLINE   98130629
REFERENCE   2  (bases 1 to 663)
  AUTHORS   Moser,B.
  TITLE     Direct Submission
  JOURNAL   Submitted (05-NOV-1997) Moser B., University of Bern, Theodor
            Kocher Institute, Freiestrasse 1, CH-3012 Bern, SWITZERLAND
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DEFINITION  H.sapiens Humig mRNA.
ACCESSION   X72755 S60728
NID         g311375
KEYWORDS    chemokine; cytokine; Humig gene; secreted protein.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 2545)
  AUTHORS   Farber,J.M.
  TITLE     Direct Submission
  JOURNAL   Submitted (22-MAR-1993) J.M. Farber, Johns Hopkins Univ. School
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of
            Medicine, Ross 1147, 720 Rutland Avenue, Baltimore, MD 21205,
USA
REFERENCE   2  (bases 1 to 2545)
  AUTHORS   Farber,J.M.
  TITLE     HuMig: a new human member of the chemokine family of cytokines
  JOURNAL   Biochem. Biophys. Res. Commun. 192 (1), 223-230 (1993)
  MEDLINE   93236577
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LOCUS       HSHUMIG      2545 bp    RNA             PRI       16-NOV-1993
DEFINITION  H.sapiens Humig mRNA.
ACCESSION   X72755 S60728
NID         g311375
KEYWORDS    chemokine; cytokine; Humig gene; secreted protein.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 2545)
  AUTHORS   Farber,J.M.
  TITLE     Direct Submission
  JOURNAL   Submitted (22-MAR-1993) J.M. Farber, Johns Hopkins Univ. School
of
            Medicine, Ross 1147, 720 Rutland Avenue, Baltimore, MD 21205,
USA
REFERENCE   2  (bases 1 to 2545)
  AUTHORS   Farber,J.M.
  TITLE     HuMig: a new human member of the chemokine family of cytokines
  JOURNAL   Biochem. Biophys. Res. Commun. 192 (1), 223-230 (1993)
  MEDLINE   93236577
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ORIGIN
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      181 gaccttaaac aatttgcccc aagcccttcc tgcgagaaaa ttgaaatcat tgctacactg
      241 aagaatggag ttcaaacatg tctaaaccca gattcagcag atgtgaagga actgattaaa
      301 aagtgggaga aacaggtcag ccaaaagaaa aagcaaaaga atgggaaaaa acatcaaaaa
      361 aagaaagttc tgaaagttcg aaaatctcaa cgttctcgtc aaaagaagac tacataagag
      421 accacttcac caataagtat tctgtgttaa aaatgttcta ttttaattat accgctatca
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      601 ttgttaaagg ctatgattgt ctttgttctt ctaccaccca ccagttgaat ttcatcatgc
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      721 ctcacaacag ctgcctggaa gagcagccct aggcttccac gtactgcagc ctccagagag
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     1021 ttccatcttg cccgctcagg ctgaccactt tatttctttt tgttcccctt tgcttcattc
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     1261 aaataaacct ttttggacac acaaattatc ttaaaactcc tgtttcactt ggttcagtac
     1321 cacatgggtg aacactcaat ggttaactaa ttcttgggtg tttatcctat ctctccaacc
     1381 agattgtcag ctccttgagg gcaagagcca cagtatattt ccctgtttct tccacagtgc
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     1681 gcacgtggta aaacacttgc ggatattctg gactgttttt aaaaaatata cagtttaccg
     1741 aaaatcatat aatcttacaa tgaaaaggac tttatagatc agccagtgac caaccttttc
     1801 ccaaccatac aaaaattcct tttcccgaag gaaaagggct ttctcaataa gcctcagctt
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     1981 tctcccatga agaaagggaa cggtgaagta ctaagcgcta gaggaagcag ccaagtcggt
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     2221 tcccacccga acgtcttatc taatcatgaa actccctagt tccttcatgt aacttccctg
     2281 aaaaatctaa gtgtttcata aatttgagag tctgtgaccc acttaccttg catctcacag
     2341 gtagacagta tataactaac aaccaaagac tacatattgt cactgacaca cacgttataa
     2401 tcatttatca tatatataca tacatgcata cactctcaaa gcaaataatt tttcacttca
     2461 aaacagtatt gacttgtata ccttgtaatt tgaaatattt tctttgttaa aatagaatgg
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LOCUS       AF002985      995 bp    mRNA             PRI       01-NOV-1997
DEFINITION  Homo sapiens putative alpha chemokine (H174) mRNA, complete
cds.
ACCESSION   AF002985
NID         g2580585
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 995)
  AUTHORS   Jacobs,K.A., Collins-Racie,L.A., Colbert,M., Duckett,M.,
            Golden-Fleet,M., Kelleher,K., Kriz,R., LaVallie,E.R.,
Merberg,D.,
            Spaulding,V., Stover,J., Williamson,M.J. and McCoy,J.M.
  TITLE       A genetic selection for isolating cDNAs encoding secreted
proteins
  JOURNAL   Gene 198 (1-2), 289-296 (1997)
  MEDLINE   98036061
REFERENCE   2  (bases 1 to 995)
  AUTHORS   Jacobs,K.A., Collins-Racie,L.A., Colbert,M., Duckett,M.,
            Golden-Fleet,M., Kelleher,K., Kriz,R., LaVallie,E.R.,
Merberg,D.,
            Spaulding,V., Stover,J., Williamson,M.J. and McCoy,J.M.
  TITLE     Direct Submission
  JOURNAL   Submitted (07-MAY-1997) Genetics Institute, 87 Cambridge Park
            Drive, Cambridge, MA 02140, USA
FEATURES             Location/Qualifiers
     source          1..995
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
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      121 gtgatattgt gtgctacagt tgttcaaggc ttccccatgt tcaaaagagg acgctgtctt
      181 tgcataggcc ctggggtaaa agcagtgaaa gtggcagata ttgagaaagc ctccataatg
      241 tacccaagta acaactgtga caaaatagaa gtgattatta ccctgaaaga aaataaagga
      301 caacgatgcc taaatcccaa atcgaagcaa gcaaggctta taatcaaaaa agttgaaaga
      361 aagaattttt aaaaatatca aaacatatga agtcctggaa aagggcatct gaaaaaccta
      421 gaacaagttt aactgtgact actgaaatga caagaattct acagtaggaa actgagactt
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      541 aaggaccaaa aacagaaata cagtcttcct gaatgaatga caatcagaat tccactgccc
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      661 catcggagtt tacaaagtgc tttcacgttc ttacttgttg tattatacat tcatgcattt
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      841 ttacagtgga gacattgaca ttattactgg agtcaagccc ttataagtca aaagcaccta
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      961 aaaaaaaaaa aaaaaaaaaa aaaaaaagcg gccgc
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LOCUS       AF030514     1371 bp    mRNA            PRI       17-JUN-1998
DEFINITION  Homo sapiens interferon stimulated T-cell alpha chemoattractant
            precursor, mRNA, complete cds.
ACCESSION   AF030514
NID         g3219692
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1371)
  AUTHORS       Cole,K.E.,   Strick,C.A.,   Paradis,T.J.,   Ogborne,K.T.,
Loetscher,M.,
            Gladue,R.P.,   Lin,W.;   Boyd,J.G.,   Moser,B.,   Wood,D.E.,
Sahagan,B.G.
            and Neote,K.
  TITLE       Interferon-inducible T cell alpha chemoattractant (I-TAC): a
novel
            non-ELR CXC chemokine with potent activity on activated T cells
            through selective high affinity binding to CXCR3
  JOURNAL   J. Exp. Med. 187 (12), 2009-2021 (1998)
  MEDLINE   98290735
REFERENCE   2  (bases 1 to 1371)
  AUTHORS   Cole,K.E., Strick,C.A. and Sahagan,B.G.
  TITLE     Direct Submission
  JOURNAL     Submitted  (20-OCT-1997)  Molecular  Sciences,  Pfizer,  Inc.,
Eastern
            Point Road, Groton, CT 06340, USA
FEATURES             Location/Qualifiers
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                     /db_xref="taxon:9606"
                     /chromosome="4"
                     /cell_type="astrocytes"
     sig_peptide     70..132
     CDS             70..354
                     /note="chemokine; I-TAC"
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                     /product="interferon stimulated T-cell alpha
                     chemoattractant precursor"
                     /db_xref="PID:g3219693"

/translation="MSVKGMAIALAVILCATVVQGFPMFKRGRCLCIGPGVKAVKVAD
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                     chemoattractant"
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      121 gttgttcaag gcttccccat gttcaaaaga ggacgctgtc tttgcatagg ccctggggta
      181 aaagcagtga aagtggcaga tattgagaaa gcctccataa tgtacccaag taacaactgt
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      361 caaaacatat gaagtcctgg aaaagggcat ctgaaaaacc tagaacaagt ttaactgtga
      421 ctactgaaat gacaagaatt ctacagtagg aaactgagac ttttctatgg ttttgtgact
      481 ttcaactttt gtacagttat gtgaaggatg aaaggtgggt gaaaggacca aaaacagaaa
      541 tacagtcttc ctgaatgaat gacaatcaga attccactgc ccaaaggagt ccagcaatta
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      961 atgtagggaa acattcttat gcatcatttg gtttgtttta taaccaattc attaaatgta
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     1261 gaaaaatgtt gaaaaataag caaatgtata cctagcaatc acttttactt tttgtaattc
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LOCUS       AF030514     1371 bp    mRNA            PRI       17-JUN-1998
DEFINITION  Homo sapiens interferon stimulated T-cell alpha chemoattractant
            precursor, mRNA, complete cds.
ACCESSION   AF030514
NID         g3219692
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1371)
  AUTHORS       Cole,K.E.,    Strick,C.A.,    Paradis,T.J.,    Ogborne,K.T.,
Loetscher,M.,
            Gladue,R.P.,    Lin,W.,    Boyd,J.G.,    Moser,B.,    Wood,D.E.,
Sahagan,B.G.
            and Neote,K.
  TITLE       Interferon-inducible T cell alpha chemoattractant (I-TAC): a
novel
            non-ELR CXC chemokine with potent activity on activated T cells
            through selective high affinity binding to CXCR3
  JOURNAL   J. Exp. Med. 187 (12), 2009-2021 (1998)
  MEDLINE   98290735
REFERENCE   2  (bases 1 to 1371)
  AUTHORS   Cole,K.E., Strick,C.A. and Sahagan,B.G.
  TITLE     Direct Submission
  JOURNAL      Submitted (20-OCT-1997) Molecular Sciences, Pfizer, Inc.,
Eastern
            Point Road, Groton, CT 06340, USA
FEATURES             Location/Qualifiers
     source          1..1371
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
                     /chromosome="4"
                     /cell_type="astrocytes"
     sig_peptide     70..132
     CDS             70..354
                     /note="chemokine; I-TAC"
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                     chemoattractant"
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LOCUS       AF030514     1371 bp    mRNA            PRI       17-JUN-1998
DEFINITION  Homo sapiens interferon stimulated T-cell alpha chemoattractant
            precursor, mRNA, complete cds.
ACCESSION   AF030514
NID         g3219692
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Vertebrata; Mammalia; Eutheria;
            Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1371)
  AUTHORS      Cole,K.E.,    Strick,C.A.,    Paradis,T.J.,    Ogborne,K.T.,
Loetscher,M.,
            Gladue,R.P.,    Lin,W.,    Boyd,J.G.,    Moser,B.,    Wood,D.E.,
Sahagan,B.G.
            and Neote,K.
  TITLE       Interferon-inducible T cell alpha chemoattractant (I-TAC): a
novel
            non-ELR CXC chemokine with potent activity on activated T cells
            through selective high affinity binding to CXCR3
  JOURNAL   J. Exp. Med. 187 (12), 2009-2021 (1998)
  MEDLINE   98290735
REFERENCE   2  (bases 1 to 1371)
  AUTHORS   Cole,K.E., Strick,C.A. and Sahagan,B.G.
  TITLE     Direct Submission
  JOURNAL     Submitted  (20-OCT-1997)  Molecular  Sciences,  Pfizer,  Inc.,
Eastern
            Point Road, Groton, CT 06340, USA
FEATURES             Location/Qualifiers
     source          1..1371
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
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     CDS             70..354
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      121 gttgttcaag gcttccccat gttcaaaaga ggacgctgtc tttgcatagg ccctggggta
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      481 ttcaactttt gtacagttat gtgaaggatg aaaggtgggt gaaaggacca aaaacagaaa
      541 tacagtcttc ctgaatgaat gacaatcaga attccactgc ccaaaggagt ccagcaatta
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      961 atgtagggaa acattcttat gcatcatttg gtttgtttta taaccaattc attaaatgta
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LOCUS       HSMDNCF      1560 bp    RNA             PRI       31-MAR-1995
DEFINITION  Human mRNA for MDNCF (monocyte-derived neutrophil chemotactic
            factor).
ACCESSION   Y00787
NID         g34518
KEYWORDS    cytokine.
SOURCE    . human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1560)
  AUTHORS   Matsushima,K.
  TITLE     Direct Submission
  JOURNAL      Submitted   (03-MAY-1988)  Matsushima   K.,   National   Cancer
Institute,,
            Bldg 560, Rm 31-19, Frederick, MD 21701
REFERENCE   2  (bases 1 to 1560)
  AUTHORS        Matsushima,K.,    Morishita,K.,    Yoshimura,T.,    Lavu,S.,
Kobayashi,Y.,
            Lew,W., Appella,E., Kung,H.F., Leonard,E.J. and Oppenheim,J.J.
  TITLE     Molecular cloning of a human monocyte-derived neutrophil
            chemotactic factor (MDNCF) and the induction of MDNCF mRNA by
            interleukin 1 and tumor necrosis factor
  JOURNAL   J. Exp. Med. 167 (6), 1883-1893 (1988)
  MEDLINE   88258376
COMMENT     for overlapping sequence see M17016 - M17017.
FEATURES             Location/Qualifiers
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        1 ctccataagg cacaaacttt cagagacagc agagcacaca agcttctagg acaagagcca
       61 ggaagaaacc accggaagga accatctcac tgtgtgtaaa catgacttcc aagctggccg
      121 tggctctctt ggcagccttc ctgatttctg cagctctgtg tgaaggtgca gttttgccaa
      181 ggagtgctaa agaacttaga tgtcagtgca taaagacata ctccaaacct ttccacccca
      241 aatttatcaa agaactgaga gtgattgaga gtggaccaca ctgcgccaac acagaaatta
      301 ttgtaaagct ttctgatgga agagagctct gtctggaccc caaggaaaac tgggtgcaga
      361 gggttgtgga gaagtttttg aagagggctg agaattcata aaaaaattca ttctctgtgg
      421 tatccaagaa tcagtgaaga tgccagtgaa acttcaagca aatctacttc aacacttcat
      481 gtattgtgtg ggtctgttgt agggttgcca gatgcaatac aagattcctg gttaaatttg
      541 aatttcagta aacaatgaat agtttttcat tgtaccatga aatatccaga acatacttat
      601 atgtaaagta ttatttattt gaatctacaa aaaacaacaa ataattttta aatataagga
      661 ttttcctaga tattgcacgg gagaatatac aaatagcaaa attgggccaa gggccaagag
      721 aatatccgaa ctttaatttc aggaattgaa tgggtttgct agaatgtgat atttgaagca
      781 tcacataaaa atgatgggac aataaatttt gccataaagt caaatttagc tggaaatcct
      841 ggattttttt ctgttaaatc tggcaaccct agtctgctag ccaggatcca caagtccttg
      901 ttccactgtg ccttggtttc tcctttattt ctaagtggaa aaagtattag ccaccatctt
      961 acctcacagt gatgttgtga ggacatgtgg aagcacttta agtttttca tcataacata
     1021 aattattttc aagtgtaact tattaaccta tttattattt atgtatttat ttaagcatca
     1081 aatatttgtg caagaatttg gaaaaataga agatgaatca ttgattgaat agttataaag
     1141 atgttatagt aaatttattt tattttagat attaaatgat gttttattag ataaatttca
     1201 atcagggttt ttagattaaa caaacaaaca attgggtacc cagttaaatt ttcatttcag
     1261 atatacaaca aataattttt tagtataagt acattattgt ttatctgaaa ttttaattga
     1321 actaacaatc ctagtttgat actcccagtc ttgtcattgc cagctgtgtt ggtagtgctg
     1381 tgttgaatta cggaataatg agttagaact attaaaacag ccaaaactcc acagtcaata
     1441 ttagtaattt cttgctggtt gaaacttgtt tattatgtac aaatagattc ttataatatt
     1501 atttaaatga ctgcattttt aaatacaagg ctttatattt ttaactttaa aaaaaaccgg
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LOCUS       HSINFGER     1172 bp    RNA             PRI       21-MAR-1995
DEFINITION  Human mRNA for gamma-interferon inducible early response gene
(with
            homology to platelet proteins).
ACCESSION   X02530 M17752
NID         g33917
KEYWORDS    interferon response; signal peptide.
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1172)
  AUTHORS   Luster,A.D., Unkeless,J.C. and Ravetch,J.V.
  TITLE      Gamma-interferon transcriptionally regulates an early-response
gene
            containing homology to platelet proteins
  JOURNAL   Nature 315 (6021), 672-676 (1985)
  MEDLINE   85240552
REFERENCE   2  (bases 1 to 1172)
  AUTHORS   Luster,A.D.
  TITLE     Direct Submission
  JOURNAL   Submitted (29-JUL-1986) to the EMBL/GenBank/DDBJ databases
COMMENT     Data kindly reviewed (29-JUL-1986) by Luster A.D.
FEATURES             Location/Qualifiers
     source          1..1172
                     /organism="Homo sapiens"
                     /strain="(U 937 histiocytic lymphoma cell line)"
                     /db_xref="taxon:9606"
     misc_RNA        1
                     /note="cap site"
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     sig_peptide     67..129
                     /note="pot. signal peptide (aa-21 to -1)"
     CDS             67..363
                     /note="early response precursor polypeptide (aa-21 to
77)"
                     /codon_start=1
                     /db_xref="PID:g33918"
                     /db_xref="SWISS-PROT:P02778"

/translation="MNQTAILICCLIFLTLSGIQGVPLSRTVRCTCISISNQPVNPRS

LEKLEIIPASQFCPRVEIIATMKKKGEKRCLNPESKAIKNLLKAVSKEMSKRSP"
     mat_peptide     130..360
                     /note="mature early response polypeptide (aa 1-77)"
     old_sequence    1138..1141
                     /note="ugaa was uga in [1]"
                     /citation=[1]
     old_sequence    1146..1148
                     /note="caa was ca in [1]"
                     /citation=[1]
     misc_feature    1155..1160
                     /note="pot. polyA signal"
     polyA_site      1172
                     /note="polyA site"
BASE COUNT     384 a    231 c    208 g    349 t
ORIGIN
        1 gagacattcc tcaattgctt agacatattc tgagcctaca gcagaggaac ctccagtctc
       61 agcaccatga atcaaactgc gattctgatt tgctgcctta tctttctgac tctaagtggc
      121 attcaaggag tacctctctc tagaaccgta cgctgtacct gcatcagcat tagtaatcaa
      181 cctgttaatc caaggtcttt agaaaaactt gaaattattc ctgcaagcca attttgtcca
      241 cgtgttgaga tcattgctac aatgaaaaag aagggtgaga agagatgtct gaatccagaa
      301 tcgaaggcca tcaagaattt actgaaagca gttagcaagg aaatgtctaa aagatctcct
      361 taaaaccaga ggggagcaaa atcgatgcag tgcttccaag gatggaccac acagaggctg
      421 cctctcccat cacttcccta catggagtat atgtcaagcc ataattgttc ttagtttgca
      481 gttacactaa aaggtgacca atgatggtca ccaaatcagc tgctactact cctgtaggaa
      541 ggttaatgtt catcatccta agctattcag taataactct accctggcac tataatgtaa
      601 gctctactga ggtgctatgt tcttagtgga tgttctgacc ctgcttcaaa tatttccctc
      661 acctttccca tcttccaagg gtactaagga atctttctgc tttggggttt atcagaattc
      721 tcagaatctc aaataactaa aaggtatgca atcaaatctg ctttttaaag aatgctcttt
      781 acttcatgga cttccactgc catcctccca aggggcccaa attctttcag tggctaccta
      841 catacaattc caaacacata caggaaggta gaaatatctg aaaatgtatg tgtaagtatt
      901 cttatttaat gaaagactgt acaaagtata agtcttagat gtatatattt cctatattgt
      961 tttcagtgta catggaataa catgtaatta agtactatgt atcaatgagt aacaggaaaa
     1021 ttttaaaaat acagatagat atatgctctg catgttacat aagataaatg tgctgaatgg
     1081 ttttcaaata aaaatgaggt actctcctgg aaatattaag aaagactatc taaatgttga
     1141 aagatcaaaa ggttaataaa gtaattataa ct
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LOCUS       SYNRPF4A      225 bp    DNA             SYN       15-JUN-1989
DEFINITION  Human recombinant platelet factor 4 (PF4) gene, complete cds.
ACCESSION   M20901
NID         g209285
KEYWORDS    platelet factor; platelet factor 4.
SOURCE      Synthetic oligonucleotide DNA, clone pIN-III-ompA-2.
  ORGANISM  artificial sequence
            artificial sequence.
REFERENCE   1  (bases 1 to 225)
  AUTHORS   Barone,A.D., Ghrayeb,J., Hammerling,U., Zucker,M.B. and
            Thorbecke,G.J.
  TITLE        The expression in Escherichia coli of recombinant human
platelet
            factor 4, a protein with immunoregulatory activity
  JOURNAL   J. Biol. Chem. 263, 8710-8715 (1988)
  MEDLINE   88243725
FEATURES             Location/Qualifiers
     source          1..225
                     /organism="artificial sequence"
                     /db_xref="taxon:29278"
     CDS             <1..>225
                     /note="recombinant platelet factor 4"
                     /codon_start=2
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                    /transl_table=11
                    /db_xref="PID:g209286"

 /translation="ASMEAEEDGDLQCLCVKTTSQVRPRHITSLEVIKAGPHCPTAQL
                    IATLKDGRKICLDLQAPLYKKIIKKLLESGS"
BASE COUNT       59 a     59 c     51 g     56 t
ORIGIN      HindIII site.
        1 agcttctatg gaagctgaag aagacggtga cctgcagtgc ctgtgcgtta aaactacttc
       61 tcaggttcgt ccgcgtcata tcacttctct ggaagttatc aaagctggtc cgcattgccc
      121 gactgctcag ctgatcgcta ctctcaaaga cggtcgtaaa atctgcctgg acctgcaggc
      181 tccgctgtac aaaaaaatca tcaaaaaact gctggaatct ggatc
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LOCUS       HUMGRO       1050 bp    mRNA            PRI       11-JUN-1993
DEFINITION  Human gro (growth regulated) gene.
ACCESSION   J03561
NID         g183622
KEYWORDS    gro gene; tumor cell.
SOURCE      Human bladder tumor cell (T24) cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1050)
  AUTHORS   Anisowicz,A., Bardwell,L. and Sager,R.
  TITLE     Constitutive overexpression of a growth-regulated gene in
            transformed Chinese hamster and human cells
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 84, 7188-7192 (1987)
  MEDLINE   88041072
COMMENT     Draft entry and computer-readable sequence kindly submitted by
            R.Sager (20-NOV-1987).
FEATURES             Location/Qualifiers
     source          1..1050
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
     sig_peptide     54..140
                     /note="signal peptide (put.); putative"
     CDS             54..377
                     /note="gro protein"
                     /codon_start=1
                     /db_xref="PID:g306806"

/translation="MARAALSAAPSNPRLLRVALLLLLLVAAGRRAAGASVATELRCQ

CLQTLQGIHPKNIQSVNVKSPGPHCAQTEVIATLKNGRKACLNPASPIVKKIIEKMLN
                     SDKSN"
     mat_peptide     141..374
                     /note="gro mature protein (put.); putative"
BASE COUNT      270 a    246 c    239 g    295 t
ORIGIN      52 bp upstream of NcoI site.
        1 ctcgccagct cttccgctcc tctcacagcc gccagacccg cctgctgagc cccatggccc
       61 gcgctgctct ctccgccgcc cccagcaatc cccggctcct gcgagtggca ctgctgctcc
      121 tgctcctggt agccgctggc cggcgcgcag caggagcgtc cgtggccact gaactgcgct
      181 gccagtgctt gcagaccctg cagggaattc accccaagaa catccaaagt gtgaacgtga
      241 agtcccccgg accccactgc gcccaaaccg aagtcatagc cacactcaag aatgggcgga
      301 aagcttgcct caatcctgca tcccccatag ttaagaaaat catcgaaaag atgctgaaca
      361 gtgacaaatc caactgacca gaagggagga ggaagctcac tggtggctgt tcctgaagga
      421 ggccctgccc ttataggaac agaagaggaa agagagacac agctgcagag gccacctgga
      481 ttgtgcctaa tgtgtttgag catcgcttag gagaagtctt ctatttattt atttattcat
      541 tagttttgaa gattctatgt taatatttta ggtgtaaaat aattaagggt atgattaact
      601 ctacctgcac actgtcctat tatattcatt ctttttgaaa tgtcaacccc aagttagttc
      661 aatctggatt catatttaat ttgaaggtag aatgttttca aatgttctcc agtcattatg
      721 ttaatatttc tgaggagcct gcaacatgcc agccactgtg atagaggctg gcggatccaa
      781 gcaaatggcc aatgagatca ttgtgaaggc aggggaatgt atgtgcacat ctgttttgta
      841 actgtttaga tgaatgtcag ttgttattta ttgaaatgat ttcacagtgt gtggtcaaca
      901 tttctcatgt tgaaacttta agaactaaaa tgttctaaat atcccttgga cattttatgt
      961 ctttcttgta aggcatactg ccttgtttaa tggtagtttt acagtgtttc tggcttagaa.
     1021 caaaggggct taattattga tgttttcgga
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LOCUS       HUMGROB5     1110 bp    mRNA            PRI       07-MAR-1995
DEFINITION  Human cytokine (GRO-beta) mRNA, complete cds.
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ACCESSION   M36820
NID         g183628
KEYWORDS    cytokine.
SOURCE      Human lymphocyte, cDNA to mRNA, clone GRO-beta.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1110)
  AUTHORS   Haskill,S., Peace,A., Morris,J., Sporn,S.A., Anisowicz,A.,
            Lee,S.W., Smith,T., Martin,G., Ralph,P. and Sager,R.
  TITLE        Identification of three related human GRO genes encoding
cytokine
            functions
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (19), 7732-7736 (1990)
  MEDLINE   91017578
COMMENT       Draft entry and computer-readable sequence for [Proc. Natl.
Acad.
            Sci. U.S.A. (1990) In press] kindly submitted
            by S.Haskill, 20-JUL-1990.
FEATURES             Location/Qualifiers
     source          1..1110
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
                     /clone="GRO-beta"
                     /tissue_type="monocyte and lymphocyte"
     gene            75..398
                     /gene="GRO-beta"
     CDS             75..398
                     /gene="GRO-beta"
                     /codon_start=1
                     /product="cytokine gro-beta"
                     /db_xref="PID:g183629"

/translation="MARATLSAAPSNPRLLRVALLLLLLVAASRRAAGAPLATELRCQ

CLQTLQGIHLKNIQSVKVKSPGPHCAQTEVIATLKNGQKACLNPASPMVKKIIEKMLK
                     NGKSN"
BASE COUNT       300 a    247 c    247 g    316 t
ORIGIN
        1 gacagagccc gggccacgga gctccttgcc agctctcctc ctcgcacagc cgctcgaacc
       61 gcctgctgag ccccatggcc cgcgccacgc tctccgccgc ccccagcaat ccccggctcc
      121 tgcgggtggc gctgctgctc ctgctcctgg tggccgccag ccggcgcgca gcaggagcgc
      181 ccctggccac tgaactgcgc tgccagtgct tgcagaccct gcagggaatt cacctcaaga
      241 acatccaaag tgtgaaggtg aagtcccccg gaccccactg cgcccaaacc gaagtcatag
      301 ccacactcaa gaatgggcag aaagcttgtc tcaacccccgc atcgcccatg gttaagaaaa
      361 tcatcgaaaa gatgctgaaa aatggcaaat ccaactgacc agaaggaagg aggaagctta
      421 ttggtggctg ttcctgaagg aggccctgcc ttacaggaac agaagaggaa agagagacac
      481 agctgcagag gccacctggc ttgcgcctaa tgtgtttgag catacttagg agaagtcttc
      541 tatttattta tttatttatt tatttgtttg ttttagaaga ttctatgtta atattttatg
      601 tgtaaaataa ggttatgatt gaatctactt gcacactctc ccattatatt tattgtttat
      661 tttaggtcaa acccaagtta gttcaatcct gattcatatt taatttgaag atagaaggtt
      721 tgcagatatt ctctagtcat ttgttaatat ttcttcgtga tgacatatca catgtcagcc
      781 actgtgatag aggctgagga atccaagaaa atggccagta agatcaatgt gacggcaggg
      841 aaatgtatgt gtgtctattt tgtaactgta aagatgaatg tcagttgtta tttattgaaa
      901 tgatttcaca gtgtgtggtc aacatttctc atgttgaagc tttaagaact aaaatgttct
      961 aaatatccct tggcatttta tgtctttctt gtaagatact gccttgttta atgttaatta
     1021 tgcagtgttt ccctctgtgt tagagcagag aggtttcgat atttattgat gttttcacaa
     1081 agaacaggaa aataaaatat ttaaaaatat
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LOCUS       HUMGROGS     1064 bp    mRNA            PRI       07-MAR-1995
DEFINITION  Human cytokine (GRO-gamma) mRNA, complete cds.
ACCESSION   M36821
NID         g183632
KEYWORDS    cytokine.
SOURCE      Human lymphocyte, cDNA to mRNA, clone GRO-gamma.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1064)
  AUTHORS   Haskill,S., Peace,A., Morris,J., Sporn,S.A., Anisowicz,A.,
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            Lee,S.W., Smith,T., Martin,G., Ralph,P. and Sager,R.
  TITLE        Identification of three related human GRO genes encoding
cytokine
            functions
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (19), 7732-7736 (1990)
  MEDLINE   91017578
COMMENT       Draft entry and computer-readable sequence for [Proc. Natl.
Acad.
            Sci. U.S.A. (1990) In press] kindly submitted
            by S.Haskill, 20-JUL-1990.
FEATURES             Location/Qualifiers
     source          1..1064
                     /organism="Homo sapiens"
                     /db_xref="taxon:9606"
                     /clone="GRO-gamma"
                     /tissue_type="lymphocyte and monocyte"
     gene            78..398
                     /gene="GRO-gamma"
     CDS             78..398
                     /gene="GRO-gamma"
                     /codon_start=1
                     /product="cytokine GRO-gamma"
                     /db_xref="PID:g183633"

/translation="MAHATLSAAPSNPRLLRVALLLLLLVGSRRAAGASVVTELRCQC

LQTLQGIHLKNIQSVNVRSPGPHCAQTEVIATLKNGKKACLNPASPMVQKIIEKILNK
                     GSTN"
BASE COUNT      281 a    237 c    239 g    305 t      2 others
ORIGIN
        1 cacagccggg tcgcaggcac ctccccngcc agctctcccg cattctgcac agcttcccga
       61 cgcgtctgct gagccccatg gcccacgcca cgctctccgc cgcccccagc aatccccggc
      121 tcctgcgggt ggcgctgctg ctcctgctcc tggtgggcag ccggcgcgca gcaggagcgt
      181 ccgtggtcac tgaactgcgc tgccagtgct tgcagacact gcagggaatt cacctcaaga
      241 acatccaaag tgtgaatgta aggtcccccg gaccccactg cgcccaaacc gaagtcatag
      301 ccacactcaa gaatgggaag aaagcttgtc tcaaccccgc atcccccatg gttcagaaaa
      361 tcatcgaaaa gatactgaac aaggggagca ccaactgaca ggagagaagt aagaagctta
      421 tcagcgtatc attgacactt cctgcagggt ggtccctgcc cttaccagag ctgaaaatga
      481 aaaagagaac agcagctttc tagggacagc tggaaaggga cttaatgtgt ttgactattt
      541 cttacgaggg ttctacttat ttatgtattt attttgaaa gcttgtattt taatatttta
      601 catgctgtta tttaaagatg tgagtgtgtt tcatcaaaca tagctcagtc ctgattattt
      661 aattggaata tgatgggttt taaatgtgtc attaaactaa tatttagtgg gagaccataa
      721 tgtgtcagcc accttgataa atgacagggt ggggaactgg agggtngggg gattgaaatg
      781 caagcaatta gtggatcact gttagggtaa gggaatgtat gtacacatct atttttata
      841 cttttttttt taaaaaagaa tgtcagttgt tatttattca aattatctca cattatgtgt
      901 tcaacatttt tatgctgaag tttcccttag acattttatg tcttgcttgt agggcataat
      961 gccttgttta atgtccattc tgcagcgttt ctctttccct tggaaaagag aatttatcat
     1021 tactgttaca tttgtacaaa tgacatgata ataaaagttt tatg
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LOCUS       HUMCTAP3      673 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Human connective tissue activation peptide III mRNA, complete
cds.
ACCESSION   M54995 M38441
NID         g181175
KEYWORDS       connective tissue activating peptide-III; platelet basic
protein;
            thromboglobulin.
SOURCE      Human platelet, cDNA to mRNA, clone lambda-c[1,2].
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 673)
  AUTHORS   Wenger,R.H., Wicki,A.N., Walz,A., Kieffer,N. and Clemetson,K.J.
  TITLE     Cloning of cDNA coding for connective tissue activating peptide
III
            from a human platelet-derived lambda gt11 expression library
  JOURNAL   Blood 73 (6), 1498-1503 (1989)
  MEDLINE   89229374
FEATURES             Location/Qualifiers
     source          1..673
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                     /db_xref="taxon:9606"
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                     /clone="lambda-c1"
                     /cell_type="platelet"
                     /tissue_type="blood"
                     /tissue_lib="lambda-gt11"
                     /map="4p13-q21"
     gene            67..453
                     /gene="PPBP"
     sig_peptide     67..168
                     /gene="PPBP"
                     /note="G00-127-391"
     CDS             67..453
                     /gene="PPBP"
                     /codon_start=1
                     /db_xref="GDB:G00-127-391"
                     /product="connective tissue activating peptide III"
                     /db_xref="PID:g181176"

/translation="MSLRLDTTPSCNSARPLHALQVLLLLSLLLTALASSTKGQTKRN

LAKGKEESLDSDLYAELRCMCIKTTSGIHPKNIQSLEVIGKGTHCNQVEVIATLKDGR
                     KICLDPDAPRIKKIVQKKLAGDESAD"
     mat_peptide     196..450
                     /gene="PPBP"
                     /note="G00-127-391"
                     /product="connective tissue activating peptide III"
     mat_peptide     208..450
                     /gene="PPBP"
                     /note="G00-127-391"
                     /product="beta-thromboglobulin"
     polyA_site      673
                     /gene="PPBP"
                     /note="G00-127-391"
BASE COUNT      202 a    149 c    139 g    183 t
ORIGIN
        1 gggcaactca ccctcactca gaggtcttct ggttctggaa acaactctag ctcagccttc
       61 tccaccatga gcctcagact tgataccacc ccttcctgta acagtgcgag accacttcat
      121 gccttgcagg tgctgctgct tctgtcattg ctgctgactg ctctggcttc ctccaccaaa
      181 ggacaaacta agagaaactt ggcgaaaggc aaagaggaaa gtctagacag tgacttgtat
      241 gctgaactcc gctgcatgtg tataaagaca acctctggaa ttcatcccaa aaacatccaa
      301 agtttggaag tgatcgggaa aggaacccat tgcaaccaag tcgaagtgat agccacactg
      361 aaggatggga ggaaaatctg cctggaccca gatgctccca gaatcaagaa aattgtacag
      421 aaaaaattgg caggtgatga atctgctgat taatttgttc tgtttctgcc aaacttcttt
      481 aactcccagg aagggtagaa ttttgaaacc ttgatttttct agagttctca tttattcagg
      541 atacctattc ttactgtatt aaaatttgga tatgtgtttc attctgtctc aaaaatcaca
      601 ttttattctg agaaggttgg ttaaaagatg gcagaaagaa gatgaaaata aataagcctg
      661 gtttcaaccc tct
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LOCUS       HUMENA78A     2177 bp    DNA             PRI       31-JAN-1996
DEFINITION  Homo sapiens neutrophil-activating peptide 78 (ENA-78) gene,
            complete cds.
ACCESSION   L37036 Z46254
NID         g607030
KEYWORDS    ENA-78 gene; homologue; neutrophil-activating factor;
            neutrophil-activating peptide 78.
SOURCE      Homo sapiens DNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 2177)
  AUTHORS   Walz,A., Burgener,R., Car,B., Baggiolini,M., Kunkel,S.L. and
            Strieter,R.M.
  TITLE     Structure and neutrophil-activating properties of a novel
            inflammatory peptide (ENA-78) with homology to interleukin 8
  JOURNAL   J. Exp. Med. 174 (6), 1355-1362 (1991)
  MEDLINE   92078844
REFERENCE   2  (bases 1 to 2177)
  AUTHORS   Walz,A.
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  TITLE     Direct Submission
  JOURNAL    Submitted (14-OCT-1994) A. Walz, University of Bern, Theodor
Kocher
            Institute, Freiestr. 1, Bern, Switzerand 3012
REFERENCE   3  (bases 1 to 2177)
  AUTHORS   Corbett,M.S., Schmitt,I., Riess,O. and Walz,A.
  TITLE     Characterization of the gene for human neutrophil-activating
            peptide 78 (ENA-78)
  JOURNAL   Biochem. Biophys. Res. Commun. 205 (1), 612-617 (1994)
  MEDLINE   95091791
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DEFINITION  H.sapiens mRNA for granulocyte chemotactic protein.
ACCESSION   Y08770
NID         g1769436
KEYWORDS    cell surface receptor; CXC chemokine; GCP-2 gene; granulocyte
            chemotactic protein.
SOURCE      human.
  ORGANISM  Homo sapiens
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            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 254)
  AUTHORS      Froyen,G.,   Proost,P.,   Ronsse,I.,   Mitera,T.,   Haelens,A.,
Wuyts,A.,
            Opdenakker,G., Van Damme,J. and Billiau,A.
  TITLE       Cloning, bacterial expression and biological characterization
of
            recombinant human granulocyte chemotactic protein-2 and
            differential  expression  of  granulocyte  chemotactic  protein-2
and
            epithelial cell-derived neutrophil activating peptide-78 mRNAs
  JOURNAL   Eur. J. Biochem. 243 (3), 762-769 (1997)
  MEDLINE   97210779
REFERENCE   2  (bases 1 to 254)
  AUTHORS   Froyen,G.F.V.
  TITLE     Direct Submission
  JOURNAL      Submitted   (10-OCT-1996)  G.F.V.  Froyen,  Rega  Institute,
University
            of Leuven, Minderbroedersstraat 10, B-3000 Leuven, BELGIUM
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DEFINITION  Human DNA for SCM-1beta precursor, complete cds.
ACCESSION   D63789
NID         g1754608
KEYWORDS    SCM-1beta; SCM-1beta precursor.
SOURCE      Homo sapiens placenta DNA, clone:hg44.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;   Mammalia;   Eutheria;   Primates;   Catarrhini;
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            Homo.
REFERENCE   1  (sites)
  AUTHORS   Yoshida,T., Imai,T., Kakizaki,M., Nishimura,M. and Yoshie,O.
  TITLE     Molecular cloning of a novel C or gamma type chemokine, SCM-1
  JOURNAL   FEBS Lett. 360 (2), 155-159 (1995)
  MEDLINE   95180438
REFERENCE   2  (sites)
  AUTHORS     Yoshida,T.,  Imai,T.,  Takagi,S.,  Nishimura,M.,  Ishikawa,I.,
Yaoi,T.
            and Yoshie,O.
  TITLE     Structure and expression of two highly related genes encoding
            SCM-1/human lymphotactin
  JOURNAL   FEBS Lett. 395 (1), 82-88 (1996)
  MEDLINE   97002294
REFERENCE   3  (bases 1 to 5669)
  AUTHORS   Yoshida,T.
  JOURNAL   Unpublished (1995)
REFERENCE   4  (bases 1 to 5669)
  AUTHORS   Yoshida,T.
  TITLE     Direct Submission
  JOURNAL     Submitted (07-AUG-1995) to the DDBJ/EMBL/GenBank databases.
Tetsuya
            Yoshida,  Shionogi  Institute  for  Medical  Science;  2-5-1,
Mishima,
            Settsu,        Osaka          566,          Japan          (E-
mail:teyoshid@fl.lab.shionogi.co.jp,
            Tel:06-382-2612, Fax:06-382-2598)
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LOCUS       D63790       5660 bp    DNA             PRI       27-DEC-1996
DEFINITION  Human DNA for SCM-1alpha precursor, complete cds.
ACCESSION   D63790
NID         g1754610
KEYWORDS    SCM-1alpha precursor; SCM-1 alpha.
SOURCE      Homo sapiens placenta DNA, clone:hg40.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata;    Mammalia;     Eutheria;     Primates;     Catarrhini;
Hominidae;
            Homo.
REFERENCE   1  (sites)
  AUTHORS   Yoshida,T., Imai,T., Kakizaki,M., Nishimura,M. and Yoshie,O.
  TITLE     Molecular cloning of a novel C or gamma type chemokine, SCM-1
  JOURNAL   FEBS Lett. 360 (2), 155-159 (1995)
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  MEDLINE   95180438
REFERENCE   2  (sites)
  AUTHORS     Yoshida,T.,  Imai,T.,  Takagi,S.,  Nishimura,M.,  Ishikawa,I.,
Yaoi,T.
            and Yoshie,O.
  TITLE     Structure and expression of two highly related genes encoding
            SCM-1/human lymphotactin
  JOURNAL   FEBS Lett. 395 (1), 82-88 (1996)
  MEDLINE   97002294
REFERENCE   3  (bases 1 to 5660)
  AUTHORS   Yoshida,T.
  JOURNAL   Unpublished (1995)
REFERENCE   4  (bases 1 to 5660)
  AUTHORS   Yoshida,T.
  TITLE     Direct Submission
  JOURNAL     Submitted  (07-AUG-1995)  to  the  DDBJ/EMBL/GenBank  databases.
Tetsuya
            Yoshida,  Shionogi  Institute  for  Medical  Science;  2-5-1,
Mishima,
            Settsu,          Osaka          566,          Japan          (E-
mail:teyoshid@f1.lab.shionogi.co.jp,
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DEFINITION  Human CX3C chemokine precursor, mRNA, alternatively spliced,
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REFERENCE   1  (bases 1 to 1635)
  AUTHORS   Bazan,J.F., Bacon,K.B., Hardiman,G., Wang,W., Soo,K., Rossi,D.,
            Greaves,D.R., Zlotnik,A. and Schall,T.J.
  TITLE     A new class of membrane-bound chemokine with a CX3C motif
  JOURNAL   Nature 385 (6617), 640-644 (1997)
  MEDLINE   97177111
REFERENCE   2  (bases 1 to 1635)
  AUTHORS   Bazan,J.F., Bacon,K.B., Hardiman,G., Wang,W., Rossi,D.,
            Greaves,D.R., Zlotnik,A. and Schall,T.J.
  TITLE     Direct Submission
  JOURNAL      Submitted  (03-MAR-1997)  Molecular  Biology,  DNAX  Research
Institute,
            901 California Ave., Palo Alto, CA 94304-1104, USA
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DEFINITION  Human CX3C chemokine precursor, mRNA, alternatively spliced,
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NID         g1888522
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REFERENCE   1  (bases 1 to 3310)
  AUTHORS   Bazan,J.F., Bacon,K.B., Hardiman,G., Wang,W., Soo,K., Rossi,D.,
            Greaves,D.R., Zlotnik,A. and Schall,T.J.
  TITLE     A new class of membrane-bound chemokine with a CX3C motif
  JOURNAL   Nature 385 (6617), 640-644 (1997)
  MEDLINE   97177111
REFERENCE   2  (bases 1 to 3310)
  AUTHORS   Bazan,J.F., Bacon,K.B., Hardiman,G., Wang,W., Rossi,D.,
            Greaves,D.R., Zlotnik,A. and Schall,T.J.
  TITLE     Direct Submission
  JOURNAL      Submitted  (07-JAN-1997)  Molecular  Biology,  DNAX  Research
Institute,
            901 California Ave., Palo Alto, CA 94304-1104, USA
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LOCUS       HSU91746     1430 bp    mRNA            PRI       12-MAR-1998
DEFINITION  Homo sapiens IL-10-inducible chemokine (HCC-4) mRNA, complete
cds.
ACCESSION   U91746
NID         g2581780
KEYWORDS    .
SOURCE      human.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1430)
  AUTHORS   Hedrick,J.A., Helms,A., Gorman,D. and Zlotnik,A.
  TITLE     Identification of a novel human CC chemokine upregulated by IL-
10
  JOURNAL   Blood (1998) In press
REFERENCE   2  (bases 1 to 1430)
  AUTHORS   Hedrick,J.A., Helms,A., Gorman,D. and Zlotnik,A.
  TITLE     Direct Submission
  JOURNAL    Submitted (02-MAR-1997) Immunology, DNAX Research Institute,
901
            California Ave, Palo Alto, CA 94304, USA
FEATURES             Location/Qualifiers
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                     /db_xref="PID:g2581781"

/translation="MKVSEAALSLLVLILIITSASRSQPKVPEWVNTPSTCCLKYYEK

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BASE COUNT      401 a    351 c    293 g    385 t
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       61 tctcgcagcc agccaaaagt tcctgagtgg gtgaacaccc catccacctg ctgcctgaag
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1381 cttagtccaa aacccaagca tgcaataaat aaaactccct tatttgacaa
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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US98/26291

A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) :Please See Extra Sheet.
US CL :Please See Extra Sheet.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 424/84, 85.1, 184.1, 186.1, 188.1, 278.1; 514/2, 8, 12, 44; 530/300, 324

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

NONE

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

Please See Extra Sheet.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US 5,141, 867 A (IVANOFF et al.) 25 August 1992, see entire document.	22-32, 45-55
A	ENG et al. The Stimulatory Effects of Interleukin (IL)-12 On Hematopoiesis Are Antagonized by IL-12-induced Interferon γ In Vivo. J. Exp. Med. May 1995, Vol.181, pages 1893-1898, see entire document.	1-21, 33-44
A	ORANGE et al. Mechanism of Interleukin 12-mediated Toxicities during Experimental Viral Infections: Role of Tumor Necrosis Factor and Glucocorticoids. J. Exp. Med. March 1995, Vol.181, pages 901-914, see entire document.	1-21, 33-44

[X] Further documents are listed in the continuation of Box C. [ ] See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search 25 MARCH 1999	Date of mailing of the international search report 15 APR 1999
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer PREMA MERTZ Telephone No. (703) 308-0196

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WU et al. Receptor-mediated in Vitro Gene Transformation by a Soluble DNA Carrier System. The Journal of Biological Chemistry. 05 April 1987, Vol.252, No. 10, pages 4429-4432, see entire document.	22-32, 45-55

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US98/26291

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

Please See Extra Sheet.

1. ☒ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

A. CLASSIFICATION OF SUBJECT MATTER:
IPC (6):

C07K 14/47, 14/52; C12N 15/12, 15/19, 15/63; A61K 38/16, 38/19, 48/00

A. CLASSIFICATION OF SUBJECT MATTER:
US CL :

424/84, 85.1, 184.1, 186.1, 188.1, 278.1; 514/2, 8, 12, 44; 530/300, 324

B. FIELDS SEARCHED
Electronic data bases consulted (Name of data base and where practicable terms used):

APS, CAN ONLINE, MEDLINE, CAPLUS
search terms: chemokine, vaccination, immunogenic, antigen, HIV, efficacy, macrophage-derived chemokine, stromal cell-derived factor, monocyte chemotactic protein, composition, administration

BOX II. OBSERVATIONS WHERE UNITY OF INVENTION WAS LACKING
This ISA found multiple inventions as follows:

This application contains the following inventions or groups of inventions which are not so linked as to form a single inventive concept under PCT Rule 13.1.

Group I, claims 1-21, 33-44, drawn to a method to enhance the efficacy of a vaccine in a subject comprising administering an antigen and one or more chemokines and a composition thereto.
Group II, claims 22-32, 45-55, drawn to a method to enhance the efficacy of a vaccine in a subject comprising administering nucleic acid sequences encoding one or more antigens and nucleic acid sequences encoding one or more chemokines.

The inventions listed as Groups I-II do not relate to a single inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:
The inventions listed as Groups I-II do not relate to a single inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:
Pursuant to 37 C.F.R. § 1.475 (d), the ISA/US considers that where multiple products and processes are claimed, the main invention shall consist of the first invention of the category first mentioned in the claims and the first recited invention of each of the other categories related thereto. Accordingly, the main invention (Group I) comprises the first-recited product and method, a method to enhance the efficacy of a vaccine in a subject comprising administering an antigen and one or more chemokines and a composition thereto. Further pursuant to 37 C.F.R. § 1.475 (d), the ISA/US considers that any feature which the subsequently recited products and methods share with the main invention does not constitute a special technical feature within the meaning of PCT Rule 13.2 and that each of such products and methods accordingly defines a separate invention.